

**IN THE UNITED STATES BANKRUPTCY COURT
DISTRICT OF DELAWARE**

In re:)	
)	Chapter 11
RS FIT NW LLC,)	
)	Case No. 20-11568 (KBO)
Debtor.)	
)	(Jointly Administered)
)	
<hr style="border: 0.5px solid black;"/>		
24 HOUR FITNESS WORLDWIDE, INC.,)	
)	
Plaintiff,)	
)	
v.)	
)	
CONTINENTAL CASUALTY COMPANY;)	Adv. Pro. No. 20-51051 (KBO)
ENDURANCE AMERICAN SPECIALTY)	
INSURANCE COMPANY; STARR SURPLUS)	
LINES INSURANCE COMPANY; ALLIANZ)	
GLOBAL RISKS US INSURANCE COMPANY;)	
LIBERTY MUTUAL INSURANCE COMPANY;)	
BEAZLEY-LLOYD'S SYNDICATES 2623/623;)	
ALLIED WORLD NATIONAL ASSURANCE)	
COMPANY; QBE SPECIALTY INSURANCE)	
COMPANY; and GENERAL SECURITY)	
INDEMNITY COMPANY OF ARIZONA,)	
)	
Defendants.)	
)	
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**PROPERTY INSURER DEFENDANTS' MOTION FOR SUMMARY JUDGMENT AND
INCORPORATED MEMORANDUM OF LAW**

EXHIBIT A-10

*Declaration of Dr. Alexis Sauer-Budge and
accompanying Expert Report attached as Exhibit A*

Executed this 8th day of November, 2023.



Alexis Sauer-Budge

EXHIBIT A

Biomedical Engineering & Sciences

Exponent[®]

**Expert Report of
Dr. Alexis Sauer-Budge**

**In:
24 Hour Fitness Worldwide, Inc.**

v.

**Continental Casualty Company;
Endurance American Specialty
Insurance Company;
Starr Surplus Lines Insurance Company;
Allianz Global Risks US Insurance
Company; Liberty Mutual
Insurance Company; Beazley-Lloyd's
Syndicates 2623/623; Allied
World National Assurance
Company; QBE Specialty Insurance
Company; and General Security
Indemnity Company of Arizona**

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**Expert Report of
Dr. Alexis Sauer-Budge**

In:
24 Hour Fitness Worldwide, Inc., *et al.*
Plaintiff,

v.

**Continental Casualty Company;
Endurance American Specialty
Insurance Company;
Starr Surplus Lines Insurance Company;
Allianz Global Risks US Insurance
Company; Liberty Mutual
Insurance Company; Beazley-Lloyd's
Syndicates 2623/623; Allied
World National Assurance
Company; QBE Specialty Insurance
Company; and General Security
Indemnity Company of Arizona**

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Prepared by

A handwritten signature in blue ink, appearing to read "Alexis Sauer-Budge".

Alexis Sauer-Budge, Ph.D.
Exponent
1075 Worcester Street
Natick, MA 01760

November 23, 2022

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1. Scope of Work and Limitations

- 1.1 Exponent, Inc. (Exponent) has been retained by Continental Casualty Company, Endurance American Specialty Insurance Company, Starr Surplus Lines Insurance Company; Allianz Global Risks US Insurance Company, Liberty Mutual Insurance Company, Beazley-Lloyd's Syndicates 2623/623, Allied World National Assurance Company, QBE Specialty Insurance Company, and General Security Indemnity Company of Arizona (“Insurers”) to provide expert services related to technical aspects associated with a business interruption claim made by 24 Hour Fitness Worldwide, Inc. (“24HF”). My scope of work includes research and review of publicly available information describing viral interactions with surfaces, viral persistence in the environment, the impact of cleaning products on viruses, and methods for detecting viruses, with emphasis on SARS-CoV-2 and factors associated with the identified case. Necessarily, my present analysis is limited to information available at the date of this report.
- 1.2 This report presents my work to date in the current matter, including work performed at my direction, as well as the findings resulting from that work. The findings presented herein are made to a reasonable degree of scientific certainty based on the information currently available. I reserve the right to supplement this report and to expand or modify opinions based on the review of additional material, including material as it becomes available through ongoing testing, discovery, newly published scientific findings, and/or through any additional work or review of additional work performed by others, including 24HF’s own experts.
- 1.3 No guarantee of relevance of the described findings is expressed or implied beyond the facts of the present matter. Any reuse of this report or its findings, outside of the present confidential legal dispute between 24HF and the Insurers is made at the sole risk of the user.

2. Qualifications and Disclosures

Qualifications

- 2.1 I am currently a Senior Managing Scientist in the Biomedical Engineering and Sciences Practice at Exponent. I hold three academic degrees: (1) a Bachelor of Science in Chemistry from Stanford University, (2) a Master of Science in Chemistry from Stanford University, and (3) a Ph.D. in Biophysics from Harvard University. My Ph.D. thesis focused on the development of single-molecule DNA sequencing technologies, which has subsequently been licensed and commercialized by Oxford Nanopore Technologies. I have been active as a research scientist and product developer for more than 20 years in academia, in industry, and as a consultant with Exponent.
- 2.2 Broadly speaking, my expertise is at the interface of biology and materials. As such, I provide consulting services in the areas of chemical and microbiological contamination analysis, surface chemistry, analytical and bioanalytical chemical analysis, assay development (e.g., nucleic acid assays, immunoassays, mammalian cell-based and microbiological assays), biosensor development, and microfluidics. I have expertise in microbiology and have overseen microbiology laboratories for over twenty years. My research has included viruses, such as HIV, influenza virus, and SARS-CoV-2. As part of my work, I have studied the interaction of microbes with surfaces. Throughout the COVID-19 public health crisis, I have advised clients on the development of COVID-19 diagnostics, the effectiveness of antimicrobial surfaces and treatments, and the efficacy of various disinfection methods.
- 2.3 For over 10 years, I held an adjunct appointment in the biomedical engineering department of Boston University. I am an active participant in peer review, including having served as a standing member of the Enabling Bioanalytical and Imaging Technologies (EBIT) NIH review committee, which reviews applications on new bioanalytical tool and emerging techniques. I have published more than 35 peer-reviewed articles and am an inventor on several patents.

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Disclosures

- 2.4 My *curriculum vitae* is provided in **Appendix A** and includes a list of the publications I have authored in the last ten years.
- 2.5 My testimony list from the past four years is provided in **Appendix B**.
- 2.5 A list of items relied upon for this matter is provided in **Appendix C**.
- 2.6 Exponent has been retained by the Insurers for my services in this matter. Exponent currently charges at a rate of \$430/hour for my time. Exponent staff members with other billing rates have assisted me with this project. No portion of our compensation is dependent on the outcome of this matter.

3. Executive Summary

- 3.1 In December 2019, an outbreak of a new respiratory disease began in Wuhan, China and thereafter spread around the world. This disease, designated COVID-19 (Coronavirus disease 2019) is caused by a virus called SARS-CoV-2 (Severe Acute Respiratory Syndrome Coronavirus 2). Coronaviruses are a type of virus which is known to cause respiratory diseases such as SARS, MERS, and the common cold. Although the science related to COVID-19 rapidly evolved over the course of the pandemic, which I continue to monitor, the essential aspects of the scientific understanding pertaining to the issues on which my opinion has been sought for this case have been repeatedly verified. Specifically, the basic characteristics of SARS-CoV-2 have been demonstrated to be similar to other coronaviruses, for example, in the way that it replicates, persists in the environment, and interacts with surfaces, as well as its susceptibility to disinfectants.
- 3.2 For reasons explained further below, it is my opinion that there is no scientific basis for the assertion that SARS-CoV-2 adversely affects the surfaces or surrounding air it contacts, or that this coronavirus remains infectious after either general degradation or disinfection by one of a wide range of effective means. In other words, SARS-CoV-2 does not physically alter, change, or damage the air that surrounds respiratory droplets that contain infectious virus or the surface on which these droplets settle, thereby allowing for easy disinfection of the surface.
- 3.3 In general, viruses—unlike bacteria and fungi (molds)—are acellular, require specific animate host cells in order to replicate, do not have mechanisms for motility (or movement), and do not have the ability to infiltrate surfaces outside of the biological context of their host cells. SARS-CoV-2, like other viruses, interacts with surfaces by a process called adsorption, which is a term that describes the physical process of organic matter settling onto a solid surface. The adsorption process is driven by molecular forces between the virus and the surface, and is modulated by the liquid, virus, and surface properties. Viruses do not have a mechanism to degrade surfaces by changing the chemical or physical properties of the underlying inanimate material. Simply stated, viruses settle on

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the outer surface of solid materials, but do not penetrate or change or impact or damage the underlying material.

- 3.4 The length of time that a virus persists on a surface is dependent on a combination of biological, physical, and chemical factors pertaining to the virus and environment. Laboratory studies designed to evaluate the factors that could impact the persistence of SARS-CoV-2 in the environment have been conducted with a variety of materials and conditions. The studies have shown that SARS-CoV-2 remains infectious *on surfaces* for times on the order of hours to days under controlled laboratory conditions. However, the laboratory studies may overestimate the time SARS-CoV-2 remains infectious on surfaces in the real-world, as the studies cannot fully replicate the complexity of the real-world environment. A limited number of studies have been published that measure the infectivity of SARS-CoV-2 in real-world environments, and those have shown that either no infectious virus was recoverable or only very low amounts of infectious virus were present. Thus, available data from real-world environments supports the conclusion that infectious SARS-CoV-2 does not remain for long on inanimate materials, let alone damage or change the underlying material.
- 3.5 The ability for SARS-CoV-2 to remain infectious in respiratory droplets and aerosols in the air has also been studied. Overall, laboratory-based studies show that *aerosolized* SARS-CoV-2 can retain infectivity for a few minutes to a few hours and that the measurable infectivity decay rates vary according to testing conditions (temperature, humidity, exposure to UV-light, and aerosolized media) and testing method. Outside of the laboratory environment, a few real-world measurements of SARS-CoV-2 infectivity in aerosols have been reported. Infectious virus was only detected in a minority of the aerosols collected, suggesting that the time that SARS-CoV-2 can remain infectious in aerosols is limited, although the natural rate of loss of infectivity is not established. Furthermore, respiratory droplets only remain airborne for a limited period of time with the majority settling to surfaces in seconds to minutes within a short distance (e.g., less than 6 feet), although the specifics depend on the size of the droplet and environmental conditions, such as humidity and air flow. These findings suggest that the length of time that infectious SARS-CoV-2 can persist in the environment either on surfaces or in the air is, as a practical matter, limited.

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- 3.6 Furthermore, the findings from the above mentioned studies regarding measurements in the laboratory of infectious virus on *surfaces* from hours to days and in *aerosols* from minutes to hours is consistent with similar measurements of other respiratory viruses, such as influenza, respiratory syncytial virus (RSV), and other coronaviruses that cause the common cold. Likewise, the genetic material of these respiratory viruses are also found in places where people with those respiratory illnesses are (or have recently been) present with only a small percentage of samples being infectious. Thus, SARS-CoV-2 is similar to other respiratory viruses in the way it interacts with inanimate materials.
- 3.7 Like other coronaviruses, SARS-CoV-2 is an enveloped virus. On the so-called “hierarchy of susceptibility of human pathogens to chemical disinfectants”, enveloped viruses are listed as being among those that are the most easily disinfected from surfaces. In accordance with that general classification, studies investigating the effectiveness of different disinfectants with a wide range of chemistries have found that SARS-CoV-2 is susceptible to many disinfection methods. Furthermore, the EPA maintains a list of disinfectants for SARS-CoV-2 (List N), which as of November 12, 2022, contains 629 products. Other non-chemical disinfection methods such as heat and UV light have also been shown to be effective against SARS-CoV-2.
- 3.8 Different disinfection processes utilize a variety of mechanisms to inactivate viruses (e.g., protein denaturation, capsid destabilization, envelope disruption, and/or nucleic acid damage). In all cases, disinfection results in irreparable damage to the viral particles. Afterwards, residual inert viral components (lipids, proteins and nucleic acids) will temporarily remain on the disinfected surface; these will eventually degrade into even smaller molecules. These components are also the building blocks of microbes, plants, and animals. Thus, the same types of inert materials are also left behind on surfaces after a disinfection process in the absence of SARS-CoV-2, for example from common microbes or material of human origin such as skin cells. Accordingly, appropriately disinfected surfaces do not transmit SARS-CoV-2. Since a wide range of disinfection methods are available, the ability to disinfect materials from SARS-CoV-2 is not seen as a practical limitation, even in complex work environments with multiple types of materials and equipment.

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- 3.9 Because viruses, including SARS-CoV-2, have no physical or chemical mechanism to damage an underlying surface and the virus degrades on its own over time, as well as the fact that surfaces can be easily disinfected with a wide-range of chemical and non-chemical means, there is no scientific basis for claiming that the mere presence of a virus can physically damage property.
- 3.10 Additionally, the transient presence of SARS-CoV-2 in the air does not permanently change the air because: (1) the respiratory droplets will settle to upward facing horizontal surfaces with the majority settling within seconds to minutes, (2) viruses have no mechanism to change the molecular composition of the surrounding air, (3) the air is not static and the volume of air in a particular space is constantly being mixed with those of adjacent spaces, and (4) viruses in respiratory droplets decay over a limited period of time rendering them non-infectious.
- 3.11 Therefore, as previously mentioned and explained further in the body of this report, there is no scientific basis for the assertion that SARS-CoV-2 physically alters, changes, or damages the air that surrounds respiratory droplets that contain infectious virus or the surface on which these droplets settle.
- 3.12 Note that this Executive Summary does not itself contain all of Exponent's technical evaluations, analyses, conclusions, and recommendations of relevance to this report. Hence, this Executive Summary should be read in conjunction with the main body of this report.

4. Scientific Review and Analysis

4.1 After a brief introduction to the insured locations, the following sections describe basic information about viruses and in particular, SARS-CoV-2, the principles of viral interactions with surfaces and air, the persistence of SARS-CoV-2 and other respiratory viruses in the environment, and its susceptibility to recommended cleaning and disinfecting products. This report is presented to provide a summary of the relevant science to address allegations by Plaintiff's witnesses that the presence of SARS-CoV-2 on property results in property damage, either due to the recommendations for cleaning¹ or that were damaged as a result of cleaning.² The report by Plaintiff's expert, Dr. Mercedes Carnethon, focuses primarily on epidemiology, which is outside of my scope of work. However, to the extent that she comments on the science within my areas of expertise, I provide a response below.

A. Background on 24 Hour Fitness, Inc. (24HF)

4.2 Prior to the COVID-19 outbreak, 24HF operated 445 clubs located across the United States in 14 states, according to the complaint. Different services were offered including personal training, group exercise classes, access to fitness equipment and a "Kids' Club". Some clubs also featured basketball courts, lap pools, saunas and "Turf Zones".³ These spaces are expected to contain a wide variety of materials, such as plastics, metals, glass, wood, tiles, artificial turf as well as equipment (e.g., treadmills).⁴ Additionally, email communications between Matt Piro, Former Director of Club Operations at 24HF, and Dan Larson, Former Environmental Health & Safety Manager at 24HF, indicate that rubber, vinyl, fabric, and carpet were present at some locations.⁵

¹ Deposition of Mr. Matt Piro (Individual), dated April 27, 2022; 103:11-18.

² Deposition of Mr. Matt Piro (Individual), dated April 27, 2022; 143: 5-21; 144:3-6.

³ Complaint for Declaratory Relief. United States Bankruptcy Court, District of Delaware, dated December 21, 2020, Case No. 20-11558.

⁴ 24 Hour Fitness. 2022. Gym Experience. https://www.24hourfitness.com/gym_experience/. Accessed on 11/14/2022.

⁵ Deposition of Mr. Dan Larson (Individual), dated April 28, 2022, Case No. 20-11558, Exhibit 6.

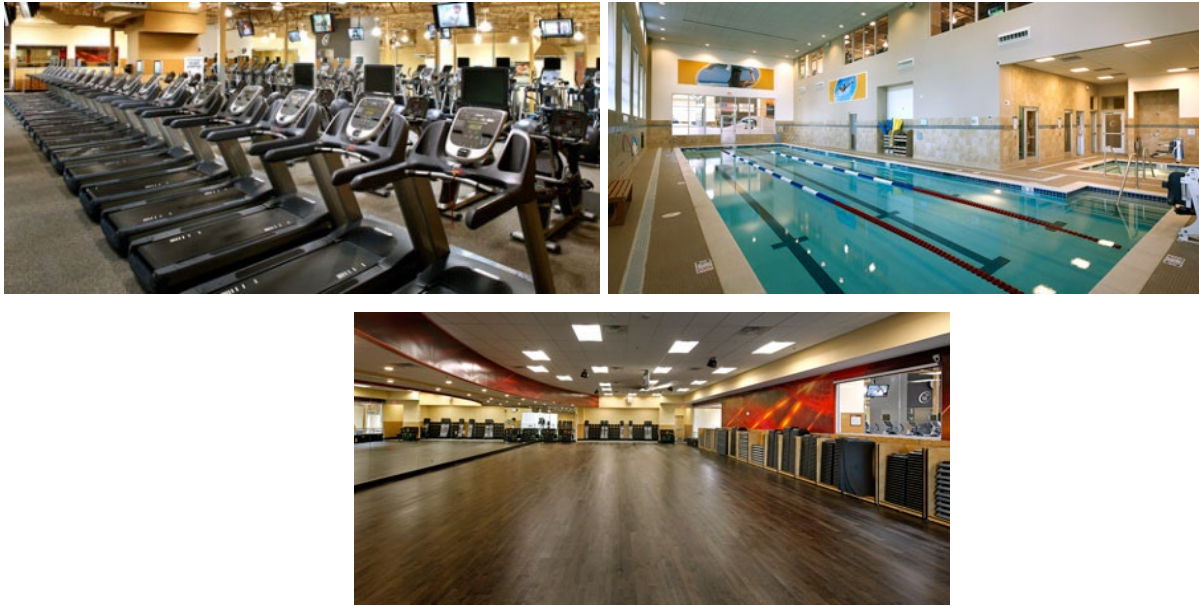


Figure 1. Images from inside 24HF Lakewood Super-Sport gym.⁶

B. COVID-19 Background

- 4.3 In December 2019, an increase in pneumonia cases in the city of Wuhan, China prompted health officials to issue an epidemiological alert.⁷ Subsequently, the virus spread worldwide and resulted in the COVID-19 pandemic. The International Committee on Taxonomy of Viruses (ICTV) announced the name of this novel pathogen as “Severe Acute Respiratory Syndrome Coronavirus 2” (SARS-CoV-2) on February 11, 2020.^{8, 9} That same

⁶ 24 Hour Fitness. 2022. Lakewood Super-Sport Gym. <https://www.24hourfitness.com/gyms/lakewood-ca/lakewood-super-sport>. Accessed on 11/14/2022.

⁷ Boni, M. F., *et al.* 2020. Evolutionary origins of the SARS-CoV-2 sarbecovirus lineage responsible for the COVID-19 pandemic. *Nat Microbiol* 5(11):1408-1417.

⁸ World Health Organization (WHO). 2020. Naming the coronavirus disease (COVID-19) and the virus that causes it. [https://www.who.int/emergencies/diseases/novel-coronavirus-2019/technical-guidance/naming-the-coronavirus-disease-\(covid-2019\)-and-the-virus-that-causes-it](https://www.who.int/emergencies/diseases/novel-coronavirus-2019/technical-guidance/naming-the-coronavirus-disease-(covid-2019)-and-the-virus-that-causes-it). Accessed on 11/12/2022.

⁹ Coronaviridae Study Group of the International Committee on Taxonomy of Viruses. 2020. The species Severe acute respiratory syndrome-related coronavirus: classifying 2019-nCoV and naming it SARS-CoV-2. *Nat Microbiol* 5(4):536-544.

day, the World Health Organization (WHO) announced “COVID-19” as the name of this new disease, which is an acronym for Corona Virus Disease 2019.¹⁰

- 4.4 By early March 2020, this novel disease had infected more than 90,000 people worldwide.¹¹ The WHO characterized COVID-19 as a pandemic on March 11, 2020.¹² To slow the spread, governments enacted various measures based, in part, on the understanding of the modes of transmission at the time. Currently, the principal transmission route is understood to be through exposure to respiratory droplets exhaled by people infected with SARS-CoV-2. The US Centers for Disease Control and Prevention (“CDC”) describes three potential routes of exposure to exhaled respiratory fluids: inhalation of aerosol particles, deposition of exhaled droplets onto exposed mucous membranes in the mouth, nose or eye (i.e., being coughed on), or touching mucous membranes with hands soiled by exhaled respiratory fluids.¹³ Thus, transmission is most likely to occur when someone is in close proximity to the infected person. The current scientific understanding is that the risk of transmission from contaminated objects (fomites) is low and fomite transmission is not a primary transmission route.^{14, 15, 16} CDC experts have confirmed that current literature evidence suggests that the risk of infection from fomites is low, and generally less than 1 in 10,000.¹⁷ Because the dominant method of

¹⁰ WHO. 2020. WHO Director-General's remarks at the media briefing on 2019-nCoV on 11 February 2020. <https://www.who.int/director-general/speeches/detail/who-director-general-s-remarks-at-the-media-briefing-on-2019-ncov-on-11-february-2020>. Accessed on 11/12/2022. Last updated on 02/11/2020.

¹¹ National Institutes of Health (NIH). 2020. Novel coronavirus structure reveals targets for vaccines and treatments. <https://www.nih.gov/news-events/nih-research-matters/novel-coronavirus-structure-reveals-targets-vaccines-treatments>. Accessed on 11/11/2022. Last updated on 03/03/2020.

¹² WHO. 2021. Listings of WHO's response to COVID-19. <https://www.who.int/news/item/29-06-2020-covidtimeline>. Accessed on 11/12/2022. Last updated on 01/29/2021.

¹³ Centers for Disease Control and Prevention (CDC). 2021. Scientific Brief: SARS-CoV-2 Transmission. <https://www.cdc.gov/coronavirus/2019-ncov/science/science-briefs/sars-cov-2-transmission.html>. Accessed on 11/11/2022. Last updated on 05/07/2021.

¹⁴ WHO. 2020. Transmission of SARS-CoV-2: implications for infection prevention precautions. <https://www.who.int/news-room/commentaries/detail/transmission-of-sars-cov-2-implications-for-infection-prevention-precautions>. Accessed on 11/12/2022. Last updated on 07/09/2020.

¹⁵ Lewis, D. 2021. COVID-19 rarely spreads through surfaces. So why are we still deep cleaning? *Nature* 590(7844):26-28.

¹⁶ Goldman, E. 2020. Exaggerated risk of transmission of COVID-19 by fomites. *Lancet Infect Dis* 20(8):892-893.

¹⁷ CDC. 2021. Science Brief: SARS-CoV-2 and Surface (Fomite) Transmission for Indoor Community Environments. <https://www.cdc.gov/coronavirus/2019-ncov/more/science-and-research/surface-transmission.html>. Accessed on 11/11/2022. Last updated on 04/05/2021.

transmission is understood to be close contact,¹⁸ the preventative strategies focused on limiting person-to-person interactions.

C. Introduction to Viruses

4.5 Microbes are abundant in our world, and commonly discussed in the context of the human environment.¹⁹ The most frequently discussed microbes in this context are bacteria, molds, and viruses, each of which are biologically distinct (see **Table 1** for a comparison). Bacteria are prokaryotic,²⁰ motile, have a diverse metabolic range, and can survive in a multitude of environments. The size of most bacteria is approximately 1 μm where μm is an abbreviation for micrometer which is one millionth of a meter²¹ (see **Figure 2** below). Molds are a type of fungi, which are eukaryotic,²² non-photosynthetic, with cell walls usually composed of chitin, and have DNA as their genetic material. Molds are multicellular, made up of long filaments, and can grow into visible colonies that range in size from approximately 10 μm to 100 cm. Fungi have the ability to extend filaments (hyphae) across distances and they can include features that allow for the manipulation of the surface they have contaminated (e.g., hyphae that can digest wood).²³ Similar to bacteria, molds can survive in many different environments.

¹⁸ Close contact is defined by the CDC as “Someone who was less than 6 feet away from an infected person (laboratory-confirmed or a clinical diagnosis) for a total of 15 minutes or more over a 24-hour period (for example, *three separate 5-minute exposures for a total of 15 minutes*). An infected person can spread the virus that causes COVID-19 starting 2 days before they have any symptoms (or, for people without symptoms, 2 days before the positive specimen collection date).” CDC. 2022. Contact Tracing Plan: Appendix A – Glossary of Key Terms. <https://www.cdc.gov/coronaviurs/2019-ncov/php/contact-tracing/contact-tracing-plan/appendix.html#contact>. Accessed on 11/12/2022. Last updated on 8/11/2022.

¹⁹ Parker, N., *et al.* 2016. Microbiology. OpenStax, Houston, TX.

²⁰ Prokaryotes are defined as organisms whose genetic material is not contained within a nucleus.

²¹ Units of length commonly used in microbiology: meter (m), decimeter (dm) (1/10 m), centimeter (cm) (1/100 m), millimeter (mm) (1/1,000 m), micrometer (μm) (1/1,000,000 m), nanometer (nm) (1/1,000,000,000 m).

²² Eukaryotic cells contain a nucleus surrounded by a nuclear membrane. Most eukaryotic cells also contain organelles, including mitochondria, Golgi apparatus, endoplasmic reticulum, and lysosomes.

²³ Fuhr, M. J., *et al.* 2011. Modelling the hyphal growth of the wood-decay fungus *Physisporinus vitreus*. *Fungal Biology* 115(9):919-932.

Table 1. Comparison of the characteristics of viruses, bacteria, and molds.²⁴

CHARACTERISTIC	VIRUS	BACTERIA ²⁵	FUNGI (MOLDS) ²⁶
Classification	Acellular	Prokaryotic (Unicellular organisms whose cells have no nucleus)	Eukaryotic (Unicellular or multicellular organisms with nuclei)
Size	~20 - 900 nm ²⁷	~1 µm	~10 µm – 100 cm
Replication	Obligate intracellular pathogens ²⁸	Some obligate intracellular pathogens, but many can replicate without host cells	Spores can lay dormant in the environment and then reproduce.
Mobility	None	Some have specialized structures to allow for movement, including cilia and flagella	Most produce specialized structures (hyphae) that can penetrate into substrate materials. ²⁹

4.6 Viruses are distinct from either bacteria or molds. Viruses are a type of microbe, consisting primarily of proteins and genetic material (i.e., acellular), that are considered inert unless found within a host organism.^{30, 31} The genetic material found within viruses can consist of deoxyribonucleic acid (DNA) or ribonucleic acid (RNA), but never both. Upon infection of a host (e.g., human cells, cells of other microorganisms), viruses utilize the host's genetic mechanisms to replicate and further infect other hosts. Without a host, a virus cannot replicate. Like bacteria and molds, viruses can be found in a number of environments. However, unlike bacteria and molds, viruses are not motile.

²⁴ Parker, N., *et al.* 2016. Microbiology. OpenStax, Houston, TX.

²⁵ Parker, N., *et al.* 2016. Microbiology. OpenStax, Houston, TX. p. 199.

²⁶ Parker, N., *et al.* 2016. Microbiology. OpenStax, Houston, TX. p. 221.

²⁷ Parker, N., *et al.* 2016. Microbiology. OpenStax, Houston, TX. p. 280.

²⁸ The term “obligate intracellular pathogens” means that the microbe can only replicate inside host cells.

²⁹ Parker, N., *et al.* 2016. Microbiology. OpenStax, Houston, TX. p. 221.

³⁰ Parker, N., *et al.* 2016. Microbiology. OpenStax, Houston, TX.

³¹ The technical term for a viral particle that is not associated with a host cell is virion. For simplification, in this report, I refer to both the extracellular and intracellular forms as “virus”.

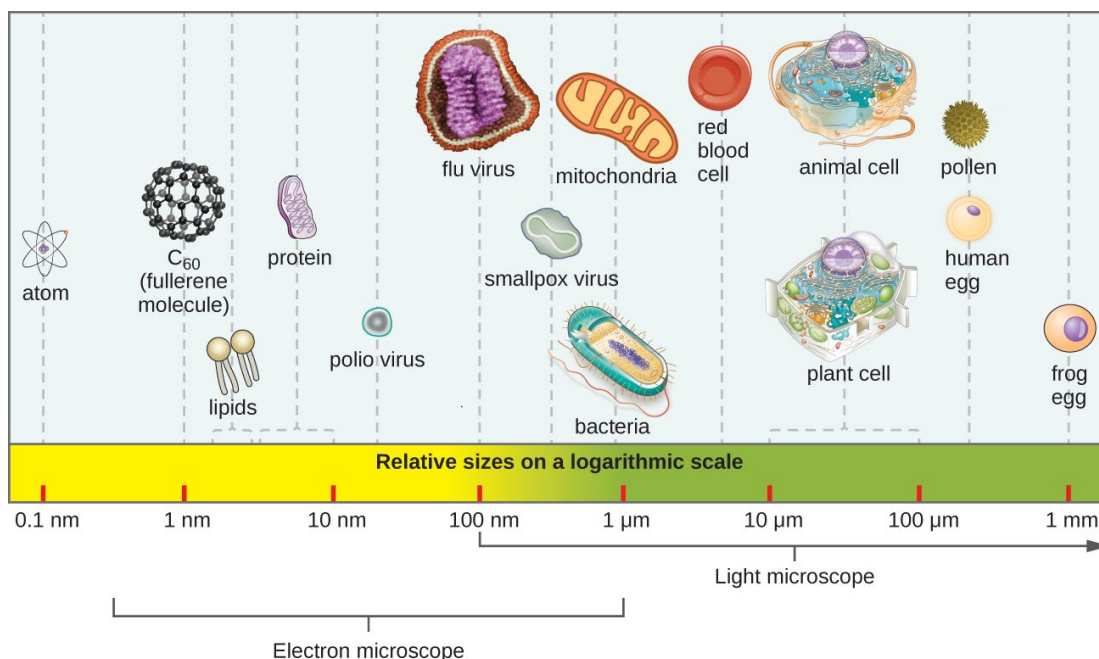


Figure 2. Illustration showing the relative sizes of different types of microbes. For references, SARS-CoV-2 is similar in size to the flu virus. Reproduced from Parker *et al.*³²

- 4.7 Of the currently known human pathogenic viruses, 21 distinct families have been classified based on differences in their structure and function.³³ Virus particles (virions) contain genetic material and associated proteins (i.e., nucleoproteins) within a protein capsid. Some viruses are “enveloped” meaning that the capsid is surrounded by a lipid bilayer. Their genetic material can contain single-stranded or double-stranded DNA or RNA, in linear or circular configuration.³⁴ The various families of human pathogenic viruses vary in shape: spherical, brick-shaped or ovoid, elongated with parallel sides and a round end, or pleomorphic (**Figure 3**).³⁵ They vary in size from approximately 20 to 900 nm (1 nm is a nanometer or one billionth of a meter).³⁶ Viruses will often contain enzymes to accompany

³² Parker, N., *et al.* 2016. Microbiology. OpenStax, Houston, TX. Figure 1.12 p. 40.

³³ Gelderblom, H. R., and S. Baron. 1996. Chapter 41: Structure and Classification of Viruses. In: Medical Microbiology. University of Texas Medical Branch at Galveston, Galveston (TX).

³⁴ RNA viruses are the most common type of virus with a heterogeneous genome structure that results in a higher mutation rate than DNA viruses. Gelderblom, H. R., and S. Baron. 1996. Chapter 41: Structure and Classification of Viruses. In: Medical Microbiology. University of Texas Medical Branch at Galveston, Galveston (TX).

³⁵ Gelderblom, H. R., and S. Baron. 1996. Chapter 41: Structure and Classification of Viruses. In: Medical Microbiology. University of Texas Medical Branch at Galveston, Galveston (TX).

³⁶ Parker, N., *et al.* 2016. Microbiology. OpenStax, Houston, TX. p. 247.

the genetic material, such as DNA-dependent RNA polymerase or protease. The enzymes found within viruses serve the primary purpose of assisting transcription (i.e., replication) of the genetic material. These enzymes by nature are highly specific and not capable of acting on anything other than the genetic material on which they were coded to act. Regardless of family, all viruses require a host to replicate, none have motility apparatus, and none act to degrade surfaces onto which they adsorb.

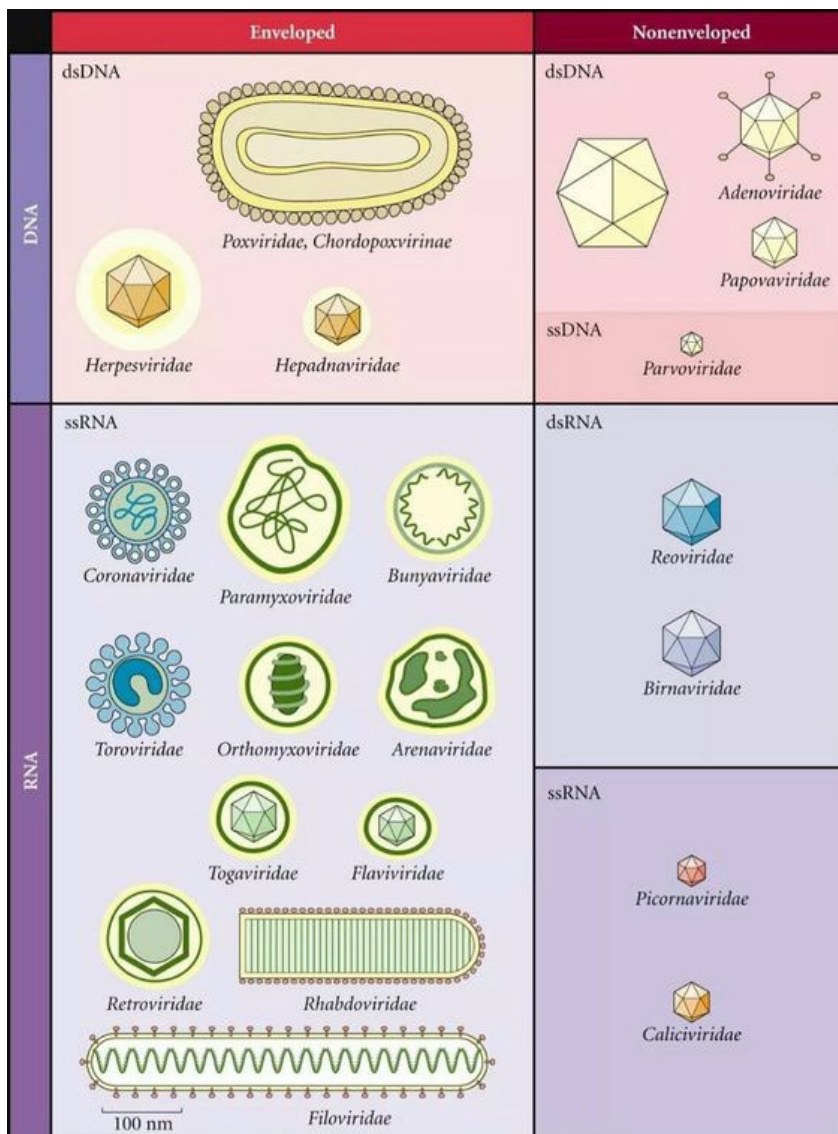


Figure 3. Illustration showing differences between various types of human pathogenic viruses. Reproduced from the Pathogen Profile Dictionary.³⁷

³⁷ Pathogen Profile Dictionary. 2022. Viruses <https://ppdictionary.com/viruses.htm>. Accessed on 11/11/2022.

- 4.8 Viruses are known to be susceptible to a variety of different disinfectant chemistries, depending in part on the structure of the virus.^{38, 39} When selecting disinfectants, it is important to consider both the virus itself, as well as the material to be disinfected, as some disinfecting chemistries can damage some materials. For example, while bleach is an excellent virus disinfectant, it would not be a good choice for a delicate silk blouse. Disinfectant chemistries will be discussed in more detail in Section G.

D. Introduction to SARS-CoV-2

General

- 4.9 SARS-CoV-2 is a coronavirus that belongs to the family *Coronaviridae*,⁴⁰ which is a known virus family and is defined by common structural features. They are spherical particles with protruding glycoproteins called spikes which resemble the shape of a crown. The envelope is composed of a lipid bilayer,⁴¹ which, as will be discussed in more detail later, makes it more susceptible to common chemical disinfectants compared to non-enveloped viruses such as rotavirus, norovirus and poliovirus.⁴² Other common coronaviruses cause mild respiratory infections in humans, such as the common cold. Examples of these closely related viruses are HCoV-229E, HCoV-NL63, HCoV-OC43 and HCoV-HKU1, which cause 15-30% of respiratory tract infections every year.⁴³ Other common respiratory viruses include adenovirus, influenza A and B, parainfluenza,

³⁸ Cimolai, N. 2020. Environmental and decontamination issues for human coronaviruses and their potential surrogates. *J Med Virol* 92(11):2498-2510.

³⁹ Artasensi, A., *et al.* 2021. Back to Basics: Choosing the Appropriate Surface Disinfectant. *Antibiotics*.

⁴⁰ Boni, M. F., *et al.* 2020. Evolutionary origins of the SARS-CoV-2 sarbecovirus lineage responsible for the COVID-19 pandemic. *Nat Microbiol* 5(11):1408-1417.

⁴¹ Tyrrell, D., and S. Myint. 1996. Coronaviruses. In: *Medical Microbiology*. S. Baron, editor. Galveston (TX): University of Texas Medical Branch at Galveston.

⁴² WHO. 2022. Coronavirus disease (COVID-19): Cleaning and disinfecting surfaces in non-health care settings. <https://www.who.int/news-room/q-a-detail/coronavirus-disease-covid-19-cleaning-and-disinfecting-surfaces-in-non-health-care-settings>. Accessed on 11/12/2022. Last updated on 03/31/2022.

⁴³ Fehr, A. R., and S. Perlman. 2015. Coronaviruses: an overview of their replication and pathogenesis. *Methods Mol Biol* 1282:1-23.

respiratory syncytial virus (RSV) A and B, rhinovirus, enterovirus and metapneumovirus.^{44, 45, 46, 47}

SARS-CoV-2 Anatomy

- 4.10 Viral proteins play an important role in the interaction with host cells and replication of their viral genome (**Figure 4**). For context, coronaviruses are 80-220 nm spherical particles with ~20 nm club-shaped spikes known as spike (S) proteins, whereas the host cells are ~10 μm (~100 times larger than the virus).⁴⁸ Plaintiff's expert, Dr. Carnethon, incorrectly states in her report that SARS-CoV-2 is "approximately 1 μm in diameter",⁴⁹ when in fact the virus has been measured to be approximately 5-10 times smaller than that.⁵⁰ The enveloped particles contain non-segmented, single-stranded positive-sense RNA (~30 kb in size).⁵¹ The major proteins composing the viral particles are nucleocapsid (N) protein (50-60 kDa) and three envelope proteins: spike (S) glycoproteins (180-220 kDa) and two transmembrane proteins (M and E, 23-35 and 9-12 kDa, respectively), which allow virion assembly.⁵²

⁴⁴ Chow, E. J., and L. A. Mermel. 2017. Hospital-Acquired Respiratory Viral Infections: Incidence, Morbidity, and Mortality in Pediatric and Adult Patients. *Open Forum Infect Dis* 4(1):ofx006.

⁴⁵ Choi, H. S., *et al.* 2017. Laboratory-based surveillance of hospital-acquired respiratory virus infection in a tertiary care hospital. *Am J Infect Control* 45(5):e45-e47.

⁴⁶ Poole, C. L., *et al.* 2019. Hospital-acquired viral respiratory infections in neonates hospitalized since birth in a tertiary neonatal intensive care unit. *J Perinatol* 39(5):683-689.

⁴⁷ Dare, R. K., and T. R. Talbot. 2016. Health Care-Acquired Viral Respiratory Diseases. *Infect Dis Clin North Am* 30(4):1053-1070.

⁴⁸ Balasuriya, U. B. R., *et al.* 2017. Chapter 24: Coronaviridae. pp. 435-461. In: Fenner's Veterinary Virology. N. J. MacLachlan and E. J. Dubovi, editors. Academic Press, Boston.

⁴⁹ Expert report of Mercedes R. Carnethon, Ph.D., October 21, 2022, Case No. 20-11558, p. 5.

⁵⁰ Tai, L., *et al.* 2021. Nanometer-resolution in situ structure of the SARS-CoV-2 postfusion spike protein. *Proc Natl Acad Sci U S A* 118(48).

⁵¹ Balasuriya, U. B. R., *et al.* 2017. Chapter 24: Coronaviridae. pp. 435-461. In: Fenner's Veterinary Virology. N. J. MacLachlan and E. J. Dubovi, editors. Academic Press, Boston.

⁵² Balasuriya, U. B. R., *et al.* 2017. Chapter 24: Coronaviridae. pp. 435-461. In: Fenner's Veterinary Virology. N. J. MacLachlan and E. J. Dubovi, editors. Academic Press, Boston.

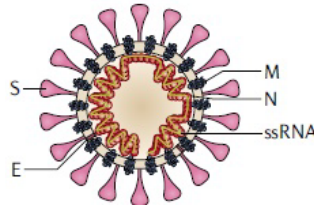


Figure 4. Illustration of coronavirus anatomy, including SARS-CoV-2, where N is the nucleoprotein, S is the spike glycoprotein, and M and E are the transmembrane proteins. Reproduced from V'kovski *et al.*⁵³

SARS-CoV-2 Replication

- 4.11 Viruses require particular types of host cells to replicate. For example, coronaviruses only infect specific cell types from humans, other mammals and avian species that express angiotensin-converting enzyme 2 (ACE2).^{54,55} If the receptor is not present on a cell, then the virus is not able to infect the cell. Additionally, if any component of the virus is damaged, the virus will not be able to undergo replication even in the presence of their host cells, e.g., if the viral proteins are denatured, the capsid is destabilized, the envelope is disrupted, or the viral RNA is damaged.

⁵³ V'Kovski, P., *et al.* 2021. Coronavirus biology and replication: implications for SARS-CoV-2. *Nat Rev Microbiol* 19(3):155-170. Figure 1 p. 2.

⁵⁴ V'Kovski, P., *et al.* 2021. Coronavirus biology and replication: implications for SARS-CoV-2. *Nat Rev Microbiol* 19(3):155-170.

⁵⁵ Ziegler, C. G. K., *et al.* 2020. SARS-CoV-2 Receptor ACE2 Is an Interferon-Stimulated Gene in Human Airway Epithelial Cells and Is Detected in Specific Cell Subsets across Tissues. *Cell* 181(5):1016-1035.e1019.

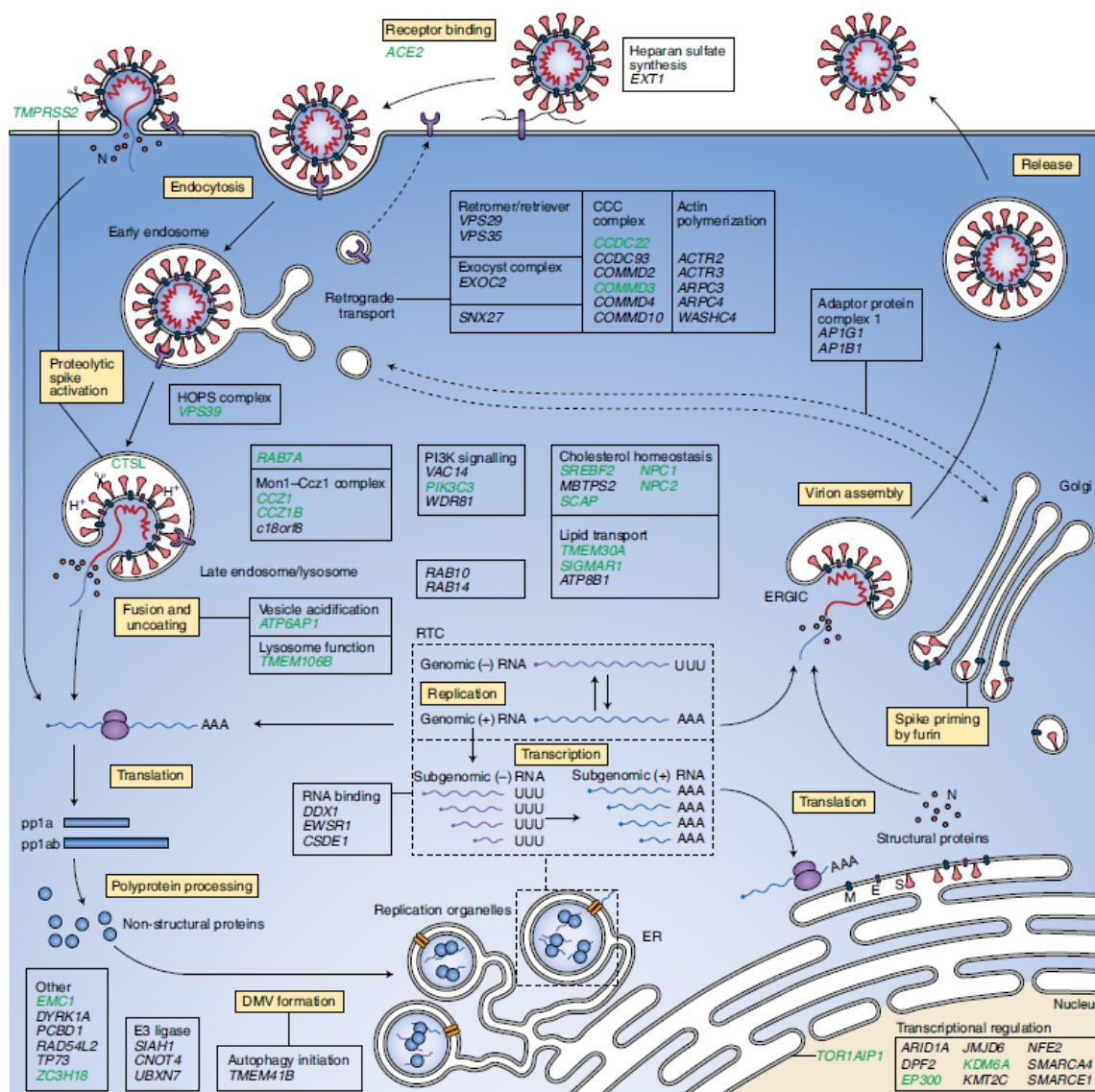


Figure 5. Illustration of coronavirus replication cycle. Reproduced from Baggen *et al.*⁵⁶

4.12 Viral replication is a complex process involving a series of specific interactions and biological contributions from both the virus and the host cell (**Figure 5**). Briefly, the virus binds to the host cell through an interaction between the S proteins and the ACE2 receptor of the host cell.⁵⁷ Then the S protein undergoes conformational rearrangements to allow

⁵⁶ Baggen, J., *et al.* 2021. Cellular host factors for SARS-CoV-2 infection. *Nature Microbiology* 6(10):1219-1232.

⁵⁷ Walls, A. C., *et al.* 2020. Structure, Function, and Antigenicity of the SARS-CoV-2 Spike Glycoprotein. *Cell* 181(2):281-292.e286.

the fusion of the viral and host membranes.^{58, 59} The SARS-CoV-2 virion is then internalized to the host cell through specific internalization pathways.^{60, 61} Following cell internalization, the virus uses the host machinery to replicate itself by initiating transcription (converting RNA to DNA) and translation (converting DNA to proteins) of the viral genetic material. A series of proteins are produced which allow the copying of the viral RNA and production of capsid proteins. These are then assembled into the viral capsid and envelope, and the genomic RNA is incorporated to the viral particle by budding from internal cell membranes. Finally, mature viral particles are secreted from the infected host cell.⁶² The complex interplay required between the virus and the host is the reason why viruses can only replicate in specific living host cells.

E. Viral Interaction with Surfaces

General

- 4.13 Outside of a biological system in which viruses bind to specific receptors on animate host cells,⁶³ SARS-CoV-2, like other viruses, interacts with inanimate surfaces by a process called adsorption, which is a term that describes the physical process of organic matter settling onto a solid surface. Adsorption can be contrasted with “absorption”, which is the process in which such organic material is taken into (absorbed into) the surface. Adsorption is described by well-developed theory that applies predictably to coronaviruses and that

⁵⁸ V'Kovski, P., *et al.* 2021. Coronavirus biology and replication: implications for SARS-CoV-2. *Nat Rev Microbiol* 19(3):155-170.

⁵⁹ Hoffmann, M., *et al.* 2020. SARS-CoV-2 Cell Entry Depends on ACE2 and TMPRSS2 and Is Blocked by a Clinically Proven Protease Inhibitor. *Cell* 181(2):271-280 e278.

⁶⁰ Walls, A. C., *et al.* 2020. Structure, Function, and Antigenicity of the SARS-CoV-2 Spike Glycoprotein. *Cell* 181(2):281-292.e286.

⁶¹ Mohan, S. V., *et al.* 2021. SARS-CoV-2 in environmental perspective: Occurrence, persistence, surveillance, inactivation and challenges. *Chem Eng J* 405:126893.

⁶² V'Kovski, P., *et al.* 2021. Coronavirus biology and replication: implications for SARS-CoV-2. *Nat Rev Microbiol* 19(3):155-170.

⁶³ As discussed in previous sections, in biological systems where a virus encounters a host cell with the appropriate receptor, then the virus can fuse with the host cell and undergo replication.

has been studied experimentally for SARS-CoV-2.^{64, 65} Computational modeling is also a useful tool for understanding the interaction between coronaviruses and surfaces, which will be discussed later in this section.

- 4.14 Since viruses do not have any mechanism to drive motion through a fluid or over a surface (as discussed above under “Introduction to Viruses”), the adsorption process is driven by the molecular forces between the virus and the surface, which are influenced by the liquid, virus, and surface properties.^{66, 67, 68, 69, 70} Moreover, viruses do not have a mechanism to degrade or damage surfaces by changing the chemical or physical properties of the substrate, for example as some fungi do with hyphae that secrete wood enzymes. Simply stated, viruses settle on the outer surface of solid materials, *but do not penetrate, alter, destroy, impact, or damage the underlying material*. Such surfaces are unchanged by viruses, including SARS-CoV-2. Importantly, the characteristics of both the environment and the virus can modulate the adsorption process through strengthening or inhibiting the molecular forces that drive viral adsorption to a particular surface.

Adsorption

- 4.15 At the molecular level, there are different types of forces that allow molecules or surfaces to interact reversibly with each other. Viral adsorption has been studied and the interaction between viruses and surfaces is generally understood to be governed by a combination of

⁶⁴ Xie, L., *et al.* 2020. A Nanomechanical Study on Deciphering the Stickiness of SARS-CoV-2 on Inanimate Surfaces. *ACS Appl Mater Interfaces* 12(52):58360-58368.

⁶⁵ Xin, Y., *et al.* 2021. Adsorption of SARS-CoV-2 Spike Protein S1 at Oxide Surfaces Studied by High-Speed Atomic Force Microscopy. *Adv Nanobiomed Res* 1(2):2000024.

⁶⁶ Gerba, C. P. 1984. Applied and theoretical aspects of virus adsorption to surfaces. *Adv Appl Microbiol* 30:133-168.

⁶⁷ Michen, B., and T. Graule. 2010. Isoelectric points of viruses. *J Appl Microbiol* 109(2):388-397.

⁶⁸ Cookson, J. T. 1969. Mechanism of virus adsorption on activated carbon. *Journal (American Water Works Association)* 61(1):52-56.

⁶⁹ Murray, J. Physical chemistry of virus adsorption and degradation on inorganic surfaces – Its relation to wastewater treatment. U.S. Environmental Protection Agency, Washington, D.C., EPA/600/2-80/134 (NTIS PB81112872), 1980.

⁷⁰ Zerda, K. S., *et al.* 1985. Adsorption of viruses to charge-modified silica. *Appl Environ Microbiol* 49(1):91-95.

van der Waals, electrostatic, and hydrophobic interactions.^{71, 72} Van der Waals forces are relatively weak chemical forces that result from the repulsion and attraction between two or more uncharged atoms or molecules that are close to each other. Hydrophobic forces describe the interactions between water and molecules that are not very soluble in water and are the molecular description of why water and oil do not mix. Electrostatic forces result from the repulsion and attraction of positive and negative charges. An understanding of adsorption informs an analysis of how viral interactions with various surfaces can be influenced by the characteristics of the virus, the surface, and the environmental conditions.

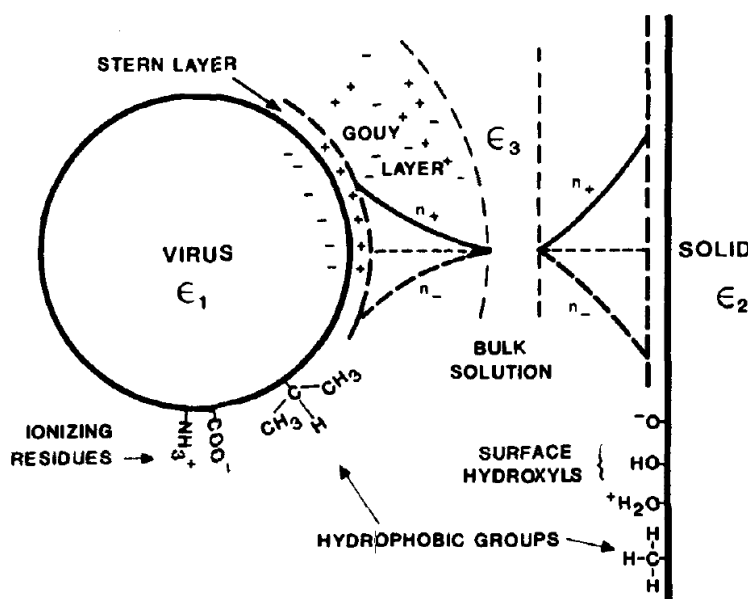


Figure 6. Diagram showing representative interaction of double layers and hydrophobic groups. Figure reproduced from Gerba.⁷³

4.16 The “DLVO (Derjaguin-Landau-Verwey-Overbeek) theory of colloid stability” is often used to describe in part the balance of electrostatic repulsion from double-layer interactions

⁷¹ Gerba, C. P. 1984. Applied and theoretical aspects of virus adsorption to surfaces. *Adv Appl Microbiol* 30:133-168.

⁷² Michen, B., and T. Graule. 2010. Isoelectric points of viruses. *J Appl Microbiol* 109(2):388-397.

⁷³ Gerba, C. P. 1984. Applied and theoretical aspects of virus adsorption to surfaces. *Adv Appl Microbiol* 30:133-168.

and the attractive van der Waals forces that describe the molecular interactions between a virus and a surface (**Figure 6**).⁷⁴ Briefly, a virus immersed in an aqueous fluid develops a fixed layer of oppositely charged ions around its surface, which in turn organizes a diffuse layer of counterions extending for some distance into the solution – this is the so-called “double layer”. The attributes of the “double layer” of ions is a function of the chemical properties of the surrounding environment (e.g., the amount of salt (ionic strength) and acid (pH) of the solution). When the thickness of this layer is reduced, the viral interaction with the surface is facilitated and allows for van der Waals forces to have an effect.⁷⁵ For enveloped viruses, such as coronaviruses, hydrophobic interactions are expected to also play an important role in their adsorption, which is influenced by the material to which it is being adsorbed.⁷⁶ In simple terms, viruses interact with surfaces at a molecular level with relatively weak forces that cause them to stick to the surface in some cases, but do not chemically alter the underlying surface through a chemical reaction nor soak into the surface through a physical absorption process.

SARS-CoV-2 Adsorption

- 4.17 The detailed theoretical basis for the adsorption of SARS-CoV-2 onto solid surfaces at the molecular level may inform an understanding of viral surface stability in the environment. As the surface of a virus has many atoms with different charges and hydrophobic character, it requires specific computational and experimental studies to fully understand the details of the interactions. Although still an area of active research, two preliminary models have been presented in the literature to describe the interaction between SARS-CoV-2 and different surfaces. Joonaki *et al.* present a model to describe the molecular interactions of SARS-CoV-2 viral interfaces in different experimental conditions (**Figure 7**).⁷⁷ The authors discuss several factors as influencing the virus adsorption process including

⁷⁴ Gerba, C. P. 1984. Applied and theoretical aspects of virus adsorption to surfaces. *Adv Appl Microbiol* 30:133-168.

⁷⁵ Gerba, C. P. 1984. Applied and theoretical aspects of virus adsorption to surfaces. *Adv Appl Microbiol* 30:133-168.

⁷⁶ Gerba, C. P. 1984. Applied and theoretical aspects of virus adsorption to surfaces. *Adv Appl Microbiol* 30:133-168.

⁷⁷ Joonaki, E., *et al.* 2020. Surface Chemistry Can Unlock Drivers of Surface Stability of SARS-CoV-2 in a Variety of Environmental Conditions. *Chem* 6(9):2135-2146.

“surface-active moieties of the viral proteins, hydrophilic or hydrophobic characteristic of the solid surface, pH of the bulk fluid, relative humidity, and temperature of the environment.”⁷⁸ In other words, this model is described by the chemical composition of the outer viral proteins, the physical properties of the solid surface and some environmental conditions.

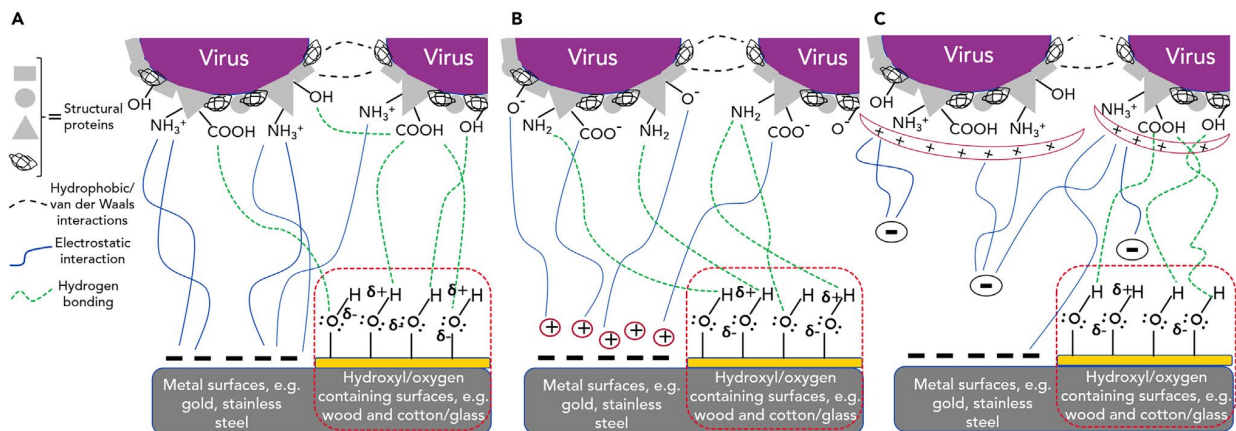


Figure 7. Model of SARS-CoV-2 adsorption to various surfaces under different environmental conditions proposed by Joonaki *et al.*⁷⁹

- 4.18 Using a different approach, researchers have used a type of computational simulation called all-atom molecular dynamics simulations to model the interactions between the SARS-CoV-2 spike glycoprotein and different surfaces (cellulose, graphite, human skin, polystyrene, polypropylene (PP), polyethylene terephthalate (PET), and polylactic acid

⁷⁸ Joonaki, E., *et al.* 2020. Surface Chemistry Can Unlock Drivers of Surface Stability of SARS-CoV-2 in a Variety of Environmental Conditions. *Chem* 6(9):2135-2146.

⁷⁹ Joonaki, E., *et al.* 2020. Surface Chemistry Can Unlock Drivers of Surface Stability of SARS-CoV-2 in a Variety of Environmental Conditions. *Chem* 6(9):2135-2146.

(PLA)).^{80, 81, 82, 83, 84} Simulation results showed that although the spike protein adsorbed initially to surfaces in a similar way, the conformation of the spike protein differed in the way it changed over time for different surfaces. For example, the spike protein remained in a similar configuration throughout the simulation with the cellulose surface, while the graphite surface induced a substantial deformation of the spike protein (**Figure 8**). While this model is a simplification of the way in which SARS-CoV-2 interacts with surfaces, it provides a visualization of the molecular interactions of adsorption to a surface and how different types of surfaces can change the molecular details of how the virus adsorbs to the surface. Other computational studies have been published exploring the interaction of SARS-CoV-2 RNA with carbon nanoparticles⁸⁵ and a model SARS-CoV-2 virion with a special type of electrostatic fiber, and both similarly demonstrate the types of forces that influence adsorption of viruses to surfaces.⁸⁶

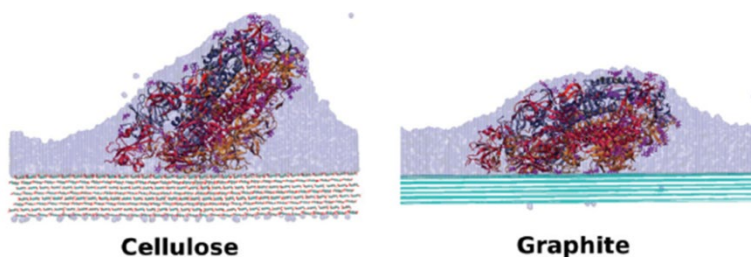


Figure 8. Snapshots of a molecular dynamics simulation of the SARS-CoV-2 spike protein adsorbing to cellulose (Left) and graphite (right). Reproduced from Marc Domingo and Jordi Faraudo 2021.⁸⁷

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- ⁸⁰ Malaspina, D. C., and J. Faraudo. 2020. Computer simulations of the interaction between SARS-CoV-2 spike glycoprotein and different surfaces. *Biointerphases* 15(5):051008.
- ⁸¹ Benkova, Z., and M. Cordeiro. 2021. Structural behavior of monomer of SARS-CoV-2 spike protein during initial stage of adsorption on graphene. *Mater Today Chem* 22:100572.
- ⁸² Domingo, M., and J. Faraudo. 2021. Interaction between SARS-CoV-2 spike glycoprotein and human skin models: a molecular dynamics study. *Soft Matter* 17(41):9457-9468.
- ⁸³ De Luca, G., *et al.* 2021. Advanced descriptors for long-range noncovalent interactions between SARS-CoV-2 spikes and polymer surfaces. *Sep Purif Technol*:120125.
- ⁸⁴ Sahihi, M., and J. Faraudo. 2022. Molecular Dynamics Simulations of Adsorption of SARS-CoV-2 Spike Protein on Polystyrene Surface. *J Chem Inf Model* 62(16):3814-3824.
- ⁸⁵ Zhang, F., *et al.* 2021. Probing nano-QSAR to assess the interactions between carbon nanoparticles and a SARS-CoV-2 RNA fragment. *Ecotoxicol Environ Saf* 219:112357.
- ⁸⁶ Javidpour, L., *et al.* 2021. Electrostatic interactions between the SARS-CoV-2 virus and a charged electret fibre. *Soft Matter* 17(16):4296-4303.
- ⁸⁷ Domingo, M., and J. Faraudo. 2021. Interaction between SARS-CoV-2 spike glycoprotein and human skin models: a molecular dynamics study. *Soft Matter* 17(41):9457-9468.

- 4.19 Additionally, two studies have been published that experimentally measure the interaction forces between the SARS-CoV-2 spike protein and various surfaces.^{88, 89} These studies used a technique called atomic force microscopy (AFM) to measure the adsorption forces. The studies confirmed that adsorption is to a greater or lesser extent governed by electrostatic forces, van der Waals interactions, and hydrophobic interactions depending on the surface characteristics and the liquid environment.

F. SARS-CoV-2 Persistence in the Environment

Introduction

- 4.20 Microbes can be found on surfaces all around us^{90, 91, 92, 93, 94, 95} (unless those surfaces have recently been disinfected). It is also well known that viruses can persist in the environment for some period of time, the length of which is dependent on a combination of biological, physical, and/or chemical factors pertaining to the virus and environment.⁹⁶ In her report, Dr. Carnethon states that “Theoretically, if these viral particles remain alive on surfaces, they can additionally infect individuals who touch these surfaces and transfer the virus to their own respiratory track through the mouth, nose or even the eyes.”⁹⁷ However, she does

⁸⁸ Xie, L., *et al.* 2020. A Nanomechanical Study on Deciphering the Stickiness of SARS-CoV-2 on Inanimate Surfaces. *ACS Appl Mater Interfaces* 12(52):58360-58368.

⁸⁹ Xin, Y., *et al.* 2021. Adsorption of SARS-CoV-2 Spike Protein S1 at Oxide Surfaces Studied by High-Speed Atomic Force Microscopy. *Adv Nanobiomed Res* 1(2):2000024.

⁹⁰ Firmesse, O., *et al.* 2012. Monitoring of bacterial load in terms of culturable and non-culturable cells on new materials placed in a delicatessen serve over counter. *International Journal of Food Microbiology* 159(3):179-185.

⁹¹ Fu, X., *et al.* 2020. Continental-Scale Microbiome Study Reveals Different Environmental Characteristics Determining Microbial Richness, Composition, and Quantity in Hotel Rooms. *mSystems* 5(3):e00119-00120.

⁹² Ross, A. A., and J. D. Neufeld. 2015. Microbial biogeography of a university campus. *Microbiome* 3(1):66.

⁹³ Lang, J. M., *et al.* 2014. The microbes we eat: abundance and taxonomy of microbes consumed in a day’s worth of meals for three diet types. *PeerJ* 2:e659.

⁹⁴ Dannemiller, K. C., *et al.* 2016. Influence of housing characteristics on bacterial and fungal communities in homes of asthmatic children. *Indoor Air* 26(2):179-192.

⁹⁵ Jeon, Y. S., *et al.* 2013. Identification of household bacterial community and analysis of species shared with human microbiome. *Curr Microbiol* 67(5):557-563.

⁹⁶ Vasickova, P., *et al.* 2010. Issues Concerning Survival of Viruses on Surfaces. *Food Environ Virol* 2(1):24-34.

⁹⁷ Expert report of Mercedes R. Carnethon, Ph.D., October 21, 2022, Case No. 20-11558, p. 20.

not discuss that viruses, including SARS-CoV-2, naturally degrade after relatively short periods of time. Perhaps more importantly, she does not discuss the lack of real-world evidence that SARS-CoV-2 can remain “alive” on surfaces for long periods of time. Dr. Carnethon’s report may give the incorrect impression that all the surfaces and all of the air at 24HF were covered or filled with infectious virus at all times. The scientific evidence does not support this point-of-view as further discussed below.

- 4.21 In this section, I discuss the science behind how viruses, in particular SARS-CoV-2, naturally degrade in the environment, including a discussion of both laboratory-based studies and the real-world evidence that SARS-CoV-2 degrades after a short period of time. The scientific studies show that even in the environment surrounding people known to be infected with COVID-19, e.g., hospital wards, only a very small to zero percentage of the environmental samples of the surfaces and air were found to have infectious virus in them, and those were found to be primarily within a very close proximity to the patient. The data supports the current understanding that COVID-19 is transmitted primarily by close contact and that the risk of fomite transmission is low, as discussed in Section B.

Measurement Methods for Virus Detection and Degradation

- 4.22 It is expected that SARS-CoV-2 will naturally degrade on its own over a relatively short period of time, depending on the environmental conditions, and that it can be cleaned from surfaces using widely available disinfectants. This understanding has been validated through laboratory studies and environmental sampling and will be summarized in the following sections, but first some background is provided as to how viruses are detected and how degradation is measured.
- 4.23 To detect specific viruses on environmental surfaces, including SARS-CoV-2, molecular tests such as polymerase chain reaction (PCR) are frequently used. These tests measure the presence of viral nucleic acid (RNA) that are extracted from the inanimate surfaces. Though PCR is a sensitive and feasible method, it does not distinguish between infectious

virus and non-infectious nucleic acids.⁹⁸ Thus, studies that use PCR to measure the presence of SARS-CoV-2 from environmental samples can only show that the virus was present, not that it is still infectious. Therefore, evidence of viral RNA persistence is not equivalent to evidence of infectious virus persistence and in studies that measure both persistence of viral RNA and infectious virus, RNA is shown to persist much longer than infectious virus.⁹⁹ Infectivity is detected using a technique called viral culture.^{100, 101} Studies on environmental surfaces use viral culture to detect whether a sample from an alleged contaminated surface contains SARS-CoV-2 that is still infectious.

Laboratory Studies of SARS-CoV-2 Activity on Surfaces over Time

- 4.24 Persistence of SARS-CoV-2 on non-porous surfaces, such as plastic, metals, glass, ceramic and rubber, and porous surfaces,¹⁰² such as cardboard, textiles, furs, faux leather, polymer

⁹⁸ Atkinson, B., and E. Petersen. 2020. SARS-CoV-2 shedding and infectivity. *Lancet* 395(10233):1339-1340.

⁹⁹ Paton, S., *et al.* 2021. Persistence of Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) Virus and Viral RNA in Relation to Surface Type and Contamination Concentration. *Appl Environ Microbiol* 87(14):e0052621.

¹⁰⁰ Viral culture is an infectivity test in which a viral solution is exposed to host cell lines. The ability of the viral solution to induce structural changes to the host cells (cytopathic effects) is measured to quantify the amount of infectious virus in a sample.

¹⁰¹ Harcourt, J., *et al.* 2020. Severe Acute Respiratory Syndrome Coronavirus 2 from Patient with Coronavirus Disease, United States. *Emerg Infect Dis* 26(6):1266-1273.

¹⁰² Owen, L., *et al.* 2022. Porous surfaces: stability and recovery of coronaviruses. *Interface Focus* 12(1):20210039.

banknotes, skin, wood, and paper, have been reported for more than a dozen laboratory conditions.^{103, 104, 105, 106, 107, 108, 109, 110, 111, 112, 113, 114, 115, 116, 117, 118, 119, 120, 121, 122, 123}

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- ¹⁰³ van Doremalen, N., *et al.* 2020. Aerosol and Surface Stability of SARS-CoV-2 as Compared with SARS-CoV-1. *N Engl J Med* 382(16):1564-1567.
- ¹⁰⁴ Chin, A. W. H., *et al.* 2020. Stability of SARS-CoV-2 in different environmental conditions. *Lancet Microbe* 1(1):e10.
- ¹⁰⁵ Harbourt, D. E., *et al.* 2020. Modeling the stability of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) on skin, currency, and clothing. *PLOS Neglected Tropical Diseases* 14(11):e0008831.
- ¹⁰⁶ Kratzel, A., *et al.* 2020. Temperature-dependent surface stability of SARS-CoV-2. *Journal of Infection* 81(3):452-482.
- ¹⁰⁷ Biryukov, J., *et al.* 2020. Increasing Temperature and Relative Humidity Accelerates Inactivation of SARS-CoV-2 on Surfaces. *mSphere* 5(4):e00441-00420.
- ¹⁰⁸ Matson, M. J., *et al.* 2020. Effect of Environmental Conditions on SARS-CoV-2 Stability in Human Nasal Mucus and Sputum. *Emerg Infect Dis* 26(9).
- ¹⁰⁹ Pastorino, B., *et al.* 2020. Prolonged Infectivity of SARS-CoV-2 in Fomites. *Emerging Infectious Disease journal* 26(9):2256.
- ¹¹⁰ Liu, Y., *et al.* Stability of SARS-CoV-2 on environmental surfaces and in human excreta. *Journal of Hospital Infection*.
- ¹¹¹ Riddell, S., *et al.* 2020. The effect of temperature on persistence of SARS-CoV-2 on common surfaces. *Virology Journal* 17(1):145.
- ¹¹² Kasloff, S. B., *et al.* 2021. Stability of SARS-CoV-2 on critical personal protective equipment. *Scientific Reports* 11(1):984.
- ¹¹³ Gidari A. 2021, SARS-CoV-2 Survival on Surfaces and the Effect of UV-C Light. *Viruses* 2021, 13, 408.
- ¹¹⁴ Ronca, S. E., *et al.* 2021. SARS-CoV-2 Viability on 16 Common Indoor Surface Finish Materials. *Herd*:1937586721991535.
- ¹¹⁵ Kwon, T., *et al.* 2021. Environmental Stability of SARS-CoV-2 on Different Types of Surfaces under Indoor and Seasonal Climate Conditions. *Pathogens* 10(2).
- ¹¹⁶ Pottage T., *et al.* 2021 A comparison of persistence of SARS-CoV-2 variants on stainless steel. *Journal of Hospital Infection*, 114 (2021) 163e166.
- ¹¹⁷ Virtanen, J., *et al.* 2021. Survival of SARS-CoV-2 on Clothing Materials. *Adv Virol* 2021:6623409.
- ¹¹⁸ Morris, D. H., *et al.* 2021. Mechanistic theory predicts the effects of temperature and humidity on inactivation of SARS-CoV-2 and other enveloped viruses. *Elife* 10.
- ¹¹⁹ Paton, S., *et al.* 2021 Persistence of Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) Virus and Viral RNA in Relation to Surface Type and Contamination Concentration. *Appl Environ Microbiol* 25;87(14):e0052621.
- ¹²⁰ Hirose, R., *et al.* 2021. Survival of Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) and Influenza Virus on Human Skin: Importance of Hand Hygiene in Coronavirus Disease 2019 (COVID-19). *Clin Infect Dis* 73(11):e4329-e4335.
- ¹²¹ Hirose R., *et al.* 2022. Stability of SARS-CoV-2 and influenza virus varies across different paper types. *Journal of Infection and Chemotherapy* 28 (2022) 252–256.
- ¹²² Magurano, F., *et al.* 2021. SARS-CoV-2 infection: the environmental endurance of the virus can be influenced by the increase of temperature. *Clin Microbiol Infect* 27(2):289.e285-289.e287.
- ¹²³ Onianwa, O., *et al.* 2022. Comparison of Surface Persistence of SARS-CoV-2 Alpha and Delta Variants on Stainless Steel at 4 degrees C and 24 degrees C. *Appl Environ Microbiol* 88(14):e0076422.

- 4.25 Overall, laboratory-based studies show that infectious SARS-CoV-2 can be recovered from surfaces for a few hours to a few days under ambient conditions and that the persistence time varies according to material and testing conditions (temperature, humidity, with or without organic load, etc.). Additionally, the stability of SARS-CoV-2 has been found to be affected by environmental conditions.^{124, 125, 126, 127, 128, 129, 130} A summary of the laboratory studies is provided in **Appendix D**. It is important to note that the experimental conditions can impact the measured persistence time and potentially lead to overestimations of persistence times in the real-world. For example, high concentrations of virus are known to protect from environmental decay.^{131, 132} Additionally, most laboratory studies do not include the various enzymes, salts, and proteins that are found in respiratory secretions and are known to inactivate viruses.¹³³
- 4.26 Although studies have shown that infectious virus can persist in lab conditions, few real-world settings measuring environmental surfaces have detected infectious virus.^{134, 135} Most environmental studies measure the presence of viral RNA rather than infectious virus, and in samples tested by viral culture, only a small proportion detect the presence of

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- ¹²⁴ Matson, M. J., *et al.* 2020. Effect of Environmental Conditions on SARS-CoV-2 Stability in Human Nasal Mucus and Sputum. *Emerg Infect Dis* 26(9).
- ¹²⁵ Guillier, L., *et al.* 2020. Modeling the Inactivation of Viruses from the Coronaviridae Family in Response to Temperature and Relative Humidity in Suspensions or on Surfaces. *Appl Environ Microbiol* 86(18):e01244-01220.
- ¹²⁶ Batejat, C., *et al.* 2021. Heat inactivation of the severe acute respiratory syndrome coronavirus 2. *J Biosaf Biosecur* 3(1):1-3.
- ¹²⁷ Raiteux, J., *et al.*, 2021. Inactivation of SARS-CoV-2 by Simulated Sunlight on Contaminated Surfaces. *Microbiol Spectr* 9(1):e0033321.
- ¹²⁸ Ratnesar-Shumate, S., *et al.* 2020. Simulated Sunlight Rapidly Inactivates SARS-CoV-2 on Surfaces. *J Infect Dis* 222(2):214-222.
- ¹²⁹ Magurano, F., *et al.* 2021. SARS-CoV-2 infection: the environmental endurance of the virus can be influenced by the increase of temperature. *Clin Microbiol Infect* 27(2):289.e285-289.e287.
- ¹³⁰ Shragai, T., *et al.* 2022. Household characteristics associated with surface contamination of SARS-CoV-2 and frequency of RT-PCR and viral culture positivity-California and Colorado, 2021. *PLoS One* 17(10):e0274946.
- ¹³¹ Bangiyev, R., *et al.* 2021. Higher Concentrations of Bacterial Enveloped Virus Phi6 Can Protect the Virus from Environmental Decay. *Appl Environ Microbiol* 87(21):e0137121.
- ¹³² Goldman, E. 2020. Exaggerated risk of transmission of COVID-19 by fomites. *Lancet Infect Dis* 20(8):892-893.
- ¹³³ Eccles, R. 2020. Respiratory mucus and persistence of virus on surfaces. *J Hosp Infect* 105(2):350.
- ¹³⁴ Meyerowitz, E. A., *et al.* 2021. Transmission of SARS-CoV-2: A Review of Viral, Host, and Environmental Factors. *Ann Intern Med* 174(1):69-79.
- ¹³⁵ Onakpoya, I. J., *et al.* 2021. SARS-CoV-2 and the role of fomite transmission: a systematic review. *F1000Res* 10(233):233.

infectious virus (**Appendix E**).^{136, 137, 138, 139, 140, 141, 142} For example, Santarpia *et al.* conducted a study of contamination on surfaces in hospitals and isolation rooms with individuals testing positive for SARS-CoV-2 where the presence of the virus would be expected.¹⁴³ The concentrations of viral genes were generally low and highly variable from sample to sample ranging from 0 to 1.75 copies/ μ L among the collected surface and aerosol samples. A subset of samples that were positive by PCR test was examined for viral culture. Cultivation of the virus was not confirmed due to the low concentration of SARS-CoV-2 recovered in these samples. Another study conducted by Zhou *et al.* also showed that though viral RNA of SARS-CoV-2 were detected on environmental surfaces and air samples, no infectious virus was cultured.¹⁴⁴ Marcenac *et al.* detected viral RNA of SARS-CoV-2 in 23 of 150 sample surfaces in households with infected members while infectious SARS-CoV-2 was only cultured in 1 of the 23 positives samples.¹⁴⁵

¹³⁶ Santarpia, J. L., *et al.* 2020. Aerosol and surface contamination of SARS-CoV-2 observed in quarantine and isolation care. *Scientific Reports* 10(1):12732.

¹³⁷ Zhou, J., *et al.* 2020. Investigating Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) Surface and Air Contamination in an Acute Healthcare Setting During the Peak of the Coronavirus Disease 2019 (COVID-19) Pandemic in London. *Clinical Infectious Diseases* 73(7):e1870-e1877.

¹³⁸ Lednicky, J. A., *et al.* 2020. Viable SARS-CoV-2 in the air of a hospital room with COVID-19 patients. *Int J Infect Dis* 100:476-482.

¹³⁹ Santarpia, J. L., *et al.* 2021. The size and culturability of patient-generated SARS-CoV-2 aerosol. *Journal of Exposure Science & Environmental Epidemiology*.

¹⁴⁰ Dumont-Leblond, N., *et al.* 2020. Low incidence of airborne SARS-CoV-2 in acute care hospital rooms with optimized ventilation. *Emerg Microbes Infect* 9(1):2597-2605.

¹⁴¹ Nissen, K., *et al.* 2020. Long-distance airborne dispersal of SARS-CoV-2 in COVID-19 wards. *Sci Rep* 10(1):19589.

¹⁴² Lednicky, J. A., *et al.* 2021. Isolation of SARS-CoV-2 from the air in a car driven by a COVID patient with mild illness. *Int J Infect Dis* 108:212-216.

¹⁴³ Santarpia, J. L., *et al.* 2020. Aerosol and surface contamination of SARS-CoV-2 observed in quarantine and isolation care. *Scientific Reports* 10(1):12732.

¹⁴⁴ Zhou, J., *et al.* 2020. Investigating SARS-CoV-2 surface and air contamination in an acute healthcare setting during the peak of the COVID-19 pandemic in London. *Clinical Infectious Diseases*.

¹⁴⁵ Marcenac., *et al.* 2021. Detection of SARS-CoV-2 on Surfaces in Households of Persons with COVID-19. *Int. J. Environ. Res. Public Health*, 18, 8184.

Degradation of Viruses on Surfaces

4.27 As discussed, in the absence of host cells, viruses naturally degrade over time on surfaces depending on the environmental conditions.¹⁴⁶ For a given virus, the underlying mechanism of degradation depends on the specifics of the environmental conditions as well as the surface.¹⁴⁷ For example SARS-CoV-2 has been shown to degrade at differing rates based on relative humidity, temperature, and evaporation.^{148, 149, 150} SARS-CoV-2 also degrades on metal surfaces, which is consistent with the findings of the interaction of other viruses with metal surfaces.¹⁵¹ Gerba describes the interaction between viruses and metallic oxides as the process in which the virus adsorbs to the metal oxide surfaces and then the RNA releases from the viral capsid and is broken down into small fragments as depicted in **Figure 9**.¹⁵² The protein-metal interaction results in degradation of intact virus over time and the virus becomes non-infectious.¹⁵³ Recent studies on SARS-CoV-2 inactivation in droplets suggest that evaporation plays a key role.^{154, 155}

¹⁴⁶ Sellaoui, L., *et al.* 2021. Make it clean, make it safe: A review on virus elimination via adsorption. *Chem Eng J* 412:128682.

¹⁴⁷ Aboubakr, H. A., *et al.* 2020. Stability of SARS-CoV-2 and other coronaviruses in the environment and on common touch surfaces and the influence of climatic conditions: A review. *Transbound Emerg Dis*.

¹⁴⁸ Grinchuk, P., 2021. Isothermal Evaporation Rate of Deposited Liquid Aerosols and the SARS-CoV-2 Coronavirus Survival. *Aerosol and Air Quality Research* 21(3).

¹⁴⁹ Lin, K., and L. C. Marr. 2020. Humidity-Dependent Decay of Viruses, but Not Bacteria, in Aerosols and Droplets Follows Disinfection Kinetics. *Environ Sci Technol* 54(2):1024-1032.

¹⁵⁰ Guillier, L., *et al.* 2020. Modeling the Inactivation of Viruses from the Coronaviridae Family in Response to Temperature and Relative Humidity in Suspensions or on Surfaces. *Appl Environ Microbiol* 86(18):e01244-01220.

¹⁵¹ Thurman, R. B., and C. P. Gerba. 1988. Characterization of the effect of aluminum metal on poliovirus. *Journal of Industrial Microbiology* 3(1):33-38.

¹⁵² Gerba, C. P. 1984. Applied and theoretical aspects of virus adsorption to surfaces. *Adv Appl Microbiol* 30:133-168.

¹⁵³ Thurman, R. B., and C. P. Gerba. 1988. Characterization of the effect of aluminum metal on poliovirus. *Journal of Industrial Microbiology* 3(1):33-38.

¹⁵⁴ Kong, Z. M., *et al.* 2022. Virus Dynamics and Decay in Evaporating Human Saliva Droplets on Fomites. *Environ Sci Technol*.

¹⁵⁵ Guo, L., *et al.* 2021. Transmission risk of viruses in large mucosalivary droplets on the surface of objects: A time-based analysis. *Infect Dis Now* 51(3):219-227.

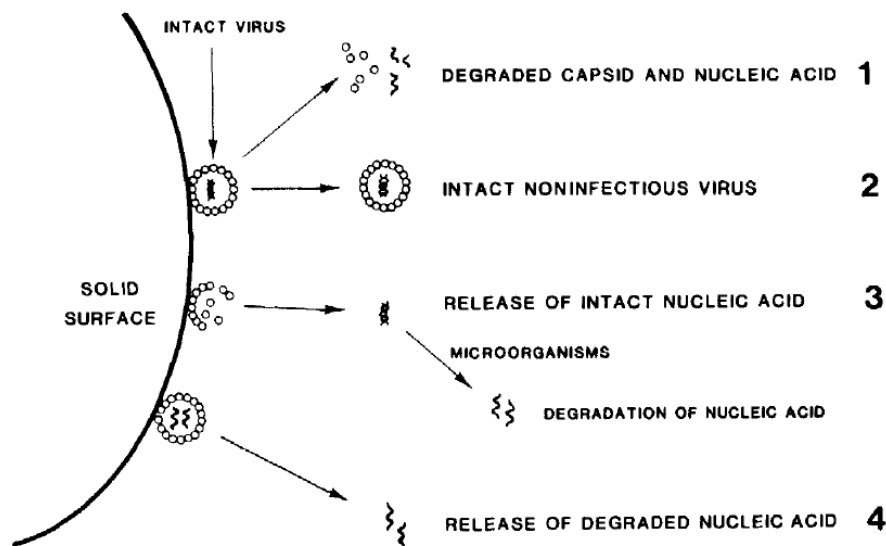


Figure 9. Mechanisms of virus inactivation on a solid surface. Reproduced from Gerba.¹⁵⁶

Laboratory Studies of SARS-CoV-2 Activity in Air over Time

4.28 The loss of infectivity of aerosolized SARS-CoV-2 over time has been measured in laboratory studies. Overall, the studies show that aerosolized SARS-CoV-2 can retain infectivity in the laboratory for a few minutes to a few hours and that the infectivity decay rate varies according to testing conditions (temperature, humidity, exposure to UV-light, and aerosolized media).^{157, 158, 159, 160, 161, 162} Additionally, the infectivity decay rate appears

¹⁵⁶ Gerba, C. P. 1984. Applied and theoretical aspects of virus adsorption to surfaces. *Adv Appl Microbiol* 30:133-168.

¹⁵⁷ van Doremalen, N., *et al.* 2020. Aerosol and Surface Stability of SARS-CoV-2 as Compared with SARS-CoV-1. *N Engl J Med* 382(16):1564-1567.

¹⁵⁸ Smither, S. J., *et al.* 2020. Experimental aerosol survival of SARS-CoV-2 in artificial saliva and tissue culture media at medium and high humidity. *Emerg Microbes Infect* 9(1):1415-1417.

¹⁵⁹ Fears, A. C., *et al.* 2020. Persistence of Severe Acute Respiratory Syndrome Coronavirus 2 in Aerosol Suspensions. *Emerg Infect Dis* 26(9).

¹⁶⁰ Schuit, M., *et al.* 2020. Airborne SARS-CoV-2 Is Rapidly Inactivated by Simulated Sunlight. *J Infect Dis* 222(4):564-571.

¹⁶¹ Oswin, H. P., *et al.* 2022. The dynamics of SARS-CoV-2 infectivity with changes in aerosol microenvironment. *Proc Natl Acad Sci U S A* 119(27):e2200109119.

¹⁶² The US Department of Homeland Security has developed a calculator that estimates the airborne decay of SARS-CoV-2 with inputs of UV index, temperature and relative humidity. Department of Homeland Security (DHS). Estimated Airborne Decay of SARS-CoV-2 (virus that causes COVID-19).

to depend strongly on the measurement technique. The majority of the measurements have been obtained using a Goldberg rotating drum. This technique has various limitations, including that it is difficult to measure the true initial infectivity level and as a result the benchmark infectivity used to compare to later time data is poorly defined.¹⁶³ A recent study used an alternative technique with improved time resolution that is based on controlled electrodynamic levitation^{164, 165, 166} to study SARS-CoV-2 infectivity in aerosols.¹⁶⁷ In this report, the infectivity was shown to reduce rapidly (in minutes) as the aerosolized droplet evaporates and comes to equilibrium with the surrounding ambient conditions.

- 4.29 Outside of the laboratory environment, a few real-world measurements of SARS-CoV-2 infectivity in aerosols have been reported (**Appendix E**). In these studies, infectious virus was collected near individuals known to have COVID-19 primarily in hospital environments,^{168, 169, 170, 171, 172, 173} although in one recently published report samples were

<https://www.dhs.gov/science-and-technology/sars-airborne-calculator>. Accessed on 11/11/2022. Last updated on 01/18/2022.

- ¹⁶³ Oswin, H. P., *et al.* 2022. The dynamics of SARS-CoV-2 infectivity with changes in aerosol microenvironment. *Proc Natl Acad Sci U S A* 119(27):e2200109119.
- ¹⁶⁴ Otero Fernandez, M., *et al.* 2020. Transformative Approach To Investigate the Microphysical Factors Influencing Airborne Transmission of Pathogens. *Appl Environ Microbiol* 86(23).
- ¹⁶⁵ Oswin, H. P., *et al.* 2021. Measuring stability of virus in aerosols under varying environmental conditions. *Aerosol Science and Technology* 55(12):1315-1320.
- ¹⁶⁶ Otero Fernandez, M., *et al.* 2019. Assessing the airborne survival of bacteria in populations of aerosol droplets with a novel technology. *J R Soc Interface* 16(150):20180779.
- ¹⁶⁷ Oswin, H. P., *et al.* 2022. The dynamics of SARS-CoV-2 infectivity with changes in aerosol microenvironment. *Proc Natl Acad Sci U S A* 119(27):e2200109119.
- ¹⁶⁸ Binder, R. A., *et al.* 2020. Environmental and Aerosolized Severe Acute Respiratory Syndrome Coronavirus 2 Among Hospitalized Coronavirus Disease 2019 Patients. *J Infect Dis* 222(11):1798-1806.
- ¹⁶⁹ Lednicky, J. A., *et al.* 2020. Viable SARS-CoV-2 in the air of a hospital room with COVID-19 patients. *Int J Infect Dis* 100:476-482.
- ¹⁷⁰ Santarpia, J. L., *et al.* 2020. Aerosol and surface contamination of SARS-CoV-2 observed in quarantine and isolation care. *Scientific Reports* 10(1):12732.
- ¹⁷¹ Santarpia, J. L., *et al.* 2021. The size and culturability of patient-generated SARS-CoV-2 aerosol. *Journal of Exposure Science & Environmental Epidemiology*.
- ¹⁷² Krambrich, J., *et al.* 2021. SARS-CoV-2 in hospital indoor environments is predominantly non-infectious. *Virology* 18(1):109.
- ¹⁷³ Lednicky, J. A., *et al.* 2020. Collection of SARS-CoV-2 Virus from the Air of a Clinic Within a University Student Health Care Center and Analyses of the Viral Genomic Sequence. *Aerosol Air Qual Res* 20(6):1167-1171.

collected from a car.¹⁷⁴ Across all of these real-world studies, infectious virus was only detected in a minority of the aerosols collected, suggesting that the time that SARS-CoV-2 can remain infectious in aerosols is limited, although the natural rate of loss of infectivity is not established. In a study conducted in 2020, air samples from a university student health care center were assessed for the presence of respiratory viruses through PCR (to measure genetic material) and through viral culture to measure infectivity. All of the samples that were positive for SARS-CoV-2 genetic material were found to be negative for infectious SARS-CoV-2. Instead, the samples were found to have infectious influenza and human coronavirus OC43 (one of the viruses which causes the common cold).¹⁷⁵ Thus, the measurement of SARS-CoV-2 genetic material in an aerosol sample cannot be equated with infectious SARS-CoV-2 virus and may in fact have other common infectious respiratory viruses in the sample.

- 4.30 Since the transmission via airborne SARS-CoV-2 is dependent on a variety of factors, including the viral load, temperature, humidity, airflow, sunlight, and host receptivity, the transient presence of respiratory droplets with infectious SARS-CoV-2 alone cannot be equated to transmissibility. As the virus decays, there is a corresponding reduction in the risk of transmission. Limited information is currently available as to the amount of active virus required to cause infection in humans and the natural infectious dose is unknown.^{176, 177} It should also be noted that the infection risk of a susceptible host is also dependent on a variety of individual and environmental factors, such as host immunity and the exposure route.¹⁷⁸

¹⁷⁴ Lednicky, J. A., *et al.* 2021. Isolation of SARS-CoV-2 from the air in a car driven by a COVID patient with mild illness. *Int J Infect Dis* 108:212-216.

¹⁷⁵ Lednicky, J. A., *et al.* 2020. Collection of SARS-CoV-2 Virus from the Air of a Clinic Within a University Student Health Care Center and Analyses of the Viral Genomic Sequence. *Aerosol Air Qual Res* 20(6):1167-1171.

¹⁷⁶ Kriegel, M., *et al.* 2022. SARS-CoV-2 Aerosol Transmission Indoors: A Closer Look at Viral Load, Infectivity, the Effectiveness of Preventive Measures and a Simple Approach for Practical Recommendations. *International Journal of Environmental Research and Public Health* 19(1):220.

¹⁷⁷ The first human SARS-CoV-2 challenge study found that 10 TCID₅₀ (equivalent to 55 FFU) delivered intranasally was sufficient to infect 53% of the participants. Killingley, B., *et al.* 2022. Safety, tolerability and viral kinetics during SARS-CoV-2 human challenge in young adults. *Nat Med*.

¹⁷⁸ Wang, C. C., *et al.* 2021. Airborne transmission of respiratory viruses. *Science* 373(6558):eabd9149.

Degradation of Viruses in Air

- 4.31 Like other respiratory viruses, the presence of SARS-CoV-2 in the air respired from a COVID-19 positive individual decays over time due to the settling of respiratory droplets out of the air and to the inactivation of the virus as the respiratory droplets evaporate. The decay of the virus can be described by first-order kinetics. The inactivation rate constant depends on a number of factors, including but not limited to the type of virus, size of respiratory droplet, temperature, humidity, exposure to UV light, and the surrounding fluid composition.¹⁷⁹ In general, larger respiratory droplets settle to upward facing surfaces rapidly (in seconds to minutes), while the smallest respiratory droplets can remain suspended for hours. For respiratory droplets that remain suspended for a longer period of time, evaporation occurs, and the virus becomes inactivated unless it infects a host cell. The rate of evaporation is dependent on multiple factors, including but not limited to, the ambient relative humidity and the composition of the respiratory droplet (e.g., salt and protein content). For respiratory droplets that are small enough to be suspended for hours, and at high relative humidity, an important factor in the likelihood of those respiratory droplets causing an infection is more heavily dependent on the amount of airflow than in situations where evaporation and subsequent inactivation happens more rapidly.
- 4.32 Due to the difficulty of studying viral decay in respiratory droplets, limited information is available on the mechanism of inactivation. Recent work with SARS-CoV-2 suggests that a key mechanism is due to efflorescence¹⁸⁰ with the respiratory droplet, a process in which the liquid evaporates and concentrates the solids (**Figure 10**).^{181, 182} Respiratory droplets are generated from inside the body, which has a high relative humidity. In general, when the droplets are respired, they enter a lower humidity environment and begin to evaporate as they come to equilibrium with the ambient conditions. As the water evaporates and the concentration of solids increases, the salts in the respiratory droplet begin to crystallize

¹⁷⁹ Wang, C. C., *et al.* 2021. Airborne transmission of respiratory viruses. *Science* 373(6558):eabd9149.

¹⁸⁰ Efflorescence is the process in which there is a spontaneous loss of water from a hydrated salt due to a change in vapor pressure. The dissolved salt migrates to the surface, then evaporates, leaving crystallized salt.

¹⁸¹ Oswin, H. P., *et al.* 2021. Measuring stability of virus in aerosols under varying environmental conditions. *Aerosol Science and Technology* 55(12):1315-1320.

¹⁸² Morris, D. H., *et al.* 2021. Mechanistic theory predicts the effects of temperature and humidity on inactivation of SARS-CoV-2 and other enveloped viruses. *Elife* 10.

(effloresce). This crystallization process and its relationship to SARS-CoV-2 inactivation was studied by cycling a suspended droplet through higher and lower relative humidity, which supported the authors' hypothesis that efflorescence may contribute to viral inactivation. Others have proposed that the limited virus survival time in droplets is governed by an increase in internal pressure within the droplet as evaporation proceeds.¹⁸³

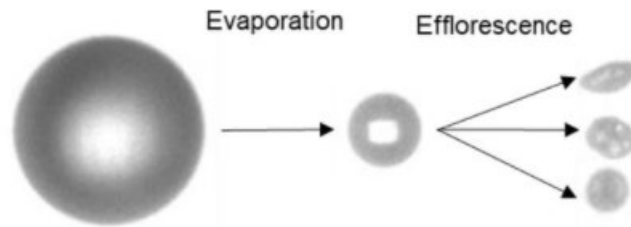


Figure 10. Illustration depicting the process of respiratory droplet desiccation over time in air. Immediately after the respiratory droplet exits the moist environment of the respiratory tract, it begins to evaporate as it comes to equilibrium with the environment. As the respiratory droplet continues to evaporate, dissolved salt moves to the surface and then crystallizes (efflorescence). Reproduced from Oswin *et al.*¹⁸⁴

Comparison to Other Respiratory Viruses

- 4.33 Other respiratory viruses have been studied and are known to have similar attributes to SARS-CoV-2 with respect to environmental persistence.¹⁸⁵ For example, laboratory-based studies of influenza viruses and human coronaviruses, which cause the common cold,¹⁸⁶ have shown the viruses can remain infectious from hours to days on different surfaces

¹⁸³ Chatterjee, S., *et al.* 2021. How coronavirus survives for hours in aerosols. *Phys Fluids* (1994) 33(8):081708.

¹⁸⁴ Oswin, H. P., *et al.* 2022. The dynamics of SARS-CoV-2 infectivity with changes in aerosol microenvironment. *Proc Natl Acad Sci U S A* 119(27):e2200109119.

¹⁸⁵ Kramer, A., and O. Assadian. 2014. Survival of Microorganisms on Inanimate Surfaces. pp. 7-26. In: *Use of Biocidal Surfaces for Reduction of Healthcare Acquired Infections*. G. Borkow, editor. Springer International Publishing, Cham.

¹⁸⁶ Centers for Disease Control and Prevention (CDC). Common Human Coronaviruses. <https://www.cdc.gov/coronavirus/downloads/Common-HCoV-fact-sheet-508.pdf> Accessed on 11/11/2022.

including steel, glass, plastic, paper, and nonwoven fabrics.^{187, 188, 189, 190, 191, 192, 193, 194, 195}

It is also known that the genetic material of respiratory viruses (which as previously discussed cannot be equated to infectious virus), such as influenza, respiratory syncytial virus (RSV), and common cold-causing coronaviruses, can be found on surfaces and in the air of crowded public places.^{196, 197, 198, 199, 200, 201} For example, Coleman *et al.* tested aerosol samples collected in Singapore's Mass Rapid Transit Network for 52 weeks during peak hours using RT-PCR. The genetic material of three respiratory viruses were identified from 89 aerosol samples. Adenovirus, RSV type A, and influenza A virus were identified in 10%, 4.5%, and 1% of the samples, respectively.²⁰² Similarly, 10% of 90 surface and air

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- ¹⁸⁷ van Doremalen, N., *et al.* 2013. Stability of Middle East respiratory syndrome coronavirus (MERS-CoV) under different environmental conditions. *Eurosurveillance* 18(38):20590.
- ¹⁸⁸ McDevitt, J., *et al.* 2010. Role of absolute humidity in the inactivation of influenza viruses on stainless steel surfaces at elevated temperatures. *Appl Environ Microbiol* 76(12):3943-3947.
- ¹⁸⁹ Dublineau, A., *et al.* 2011. Persistence of the 2009 Pandemic Influenza A (H1N1) Virus in Water and on Non-Porous Surface. *PLOS ONE* 6(11):e28043.
- ¹⁹⁰ Noyce, J. O., *et al.* 2007. Inactivation of influenza A virus on copper versus stainless steel surfaces. *Appl Environ Microbiol* 73(8):2748-2750.
- ¹⁹¹ Wood, J. P., *et al.* 2010. Environmental Persistence of a Highly Pathogenic Avian Influenza (H5N1) Virus. *Environmental Science & Technology* 44(19):7515-7520.
- ¹⁹² Kampf, G., *et al.* 2020. Persistence of coronaviruses on inanimate surfaces and their inactivation with biocidal agents. *J Hosp Infect* 104(3):246-251.
- ¹⁹³ Hirose R., *et al.* 2022. Stability of SARS-CoV-2 and influenza virus varies across different paper types. *Journal of Infection and Chemotherapy* 28 (2022) 252–256.
- ¹⁹⁴ Gidari, A., *et al.* 2021. SARS-CoV-2 Survival on Surfaces and the Effect of UV-C Light. *Viruses* 13(3).
- ¹⁹⁵ Paton, S., *et al.* 2021 Persistence of Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) Virus and Viral RNA in Relation to Surface Type and Contamination Concentration. *Appl Environ Microbiol* 25;87(14):e0052621.
- ¹⁹⁶ Ren, S. Y., *et al.* 2020. Stability and infectivity of coronaviruses in inanimate environments. *World J Clin Cases* 8(8):1391-1399.
- ¹⁹⁷ Boone, S. A., and C. P. Gerba. 2005. The occurrence of influenza A virus on household and day care center fomites. *J Infect* 51(2):103-109.
- ¹⁹⁸ Mukherjee, D. V., *et al.* 2012. Survival of influenza virus on hands and fomites in community and laboratory settings. *Am J Infect Control* 40(7):590-594.
- ¹⁹⁹ Memish, Z. A., *et al.* 2014. Environmental sampling for respiratory pathogens in Jeddah airport during the 2013 Hajj season. *Am J Infect Control* 42(12):1266-1269.
- ²⁰⁰ La Rosa, G., *et al.* 2013. Viral infections acquired indoors through airborne, droplet or contact transmission. *Ann Ist Super Sanita* 49(2):124-132.
- ²⁰¹ Hall, C. B., and R. G. Douglas, Jr. 1981. Modes of transmission of respiratory syncytial virus. *J Pediatr* 99(1):100-103.
- ²⁰² Coleman, K. K., *et al.* 2018. Bioaerosol Sampling for Respiratory Viruses in Singapore's Mass Rapid Transit Network. *Scientific Reports* 8(1):17476.

samples collected weekly from various public spaces in Finland from 2015 to 2016 tested positive for the genetic material of rhinovirus, coronavirus, adenovirus and/or influenza A.²⁰³ Studies that measured the infectivity of common respiratory viruses on environmental samples found that few, if any, samples had infectious virus on them.^{204, 205} Thus, SARS-CoV-2 is similar to other respiratory viruses in the way it interacts with inanimate materials.

- 4.34 When considering the results of environmental contamination studies, it is important to understand the methods being used and their limitations. As previously discussed above under “Measurement Methods for Virus Detection and Degradation”, PCR has been used to test for the presence of SARS-CoV-2 viral RNA on surfaces. Because it only measures the presence of viral RNA (a part of the virus) and not the presence of the whole virus nor the infectiousness of the virus, this test cannot distinguish between inactivated and infectious virus. It is also important to understand that this type of test only tests for one thing – the presence of SARS-CoV-2 viral RNA. It does not test for the presence of other viruses nor other microbes. Thus, one should not interpret SARS-CoV-2 PCR results as the absence of other microbes. This is highlighted by a recent study by Marotz *et al.* that looked at the presence of SARS-CoV-2 viral RNA and other microbes in the patients’ rooms.²⁰⁶ Viral RNA was found in the presence of a wide variety of other microbes, including from the actinobacteria, firmicutes, proteobacteria and Bacteroidetes phyla.²⁰⁷ The wide variety of microbes found in this study is consistent with another study of how a

²⁰³ Ikonen, N., *et al.* 2018. Deposition of respiratory virus pathogens on frequently touched surfaces at airports. BMC Infectious Diseases 18(1):437.

²⁰⁴ Mukherjee, D. V., *et al.* 2012. Survival of influenza virus on hands and fomites in community and laboratory settings. Am J Infect Control 40(7):590-594.

²⁰⁵ Coleman, K. K., *et al.* 2018. Bioaerosol Sampling for Respiratory Viruses in Singapore’s Mass Rapid Transit Network. Scientific Reports 8(1):17476.

²⁰⁶ Marotz, C., *et al.* 2021. SARS-CoV-2 detection status associates with bacterial community composition in patients and the hospital environment. Microbiome 9(1):132.

²⁰⁷ Phylum (plural: phyla) is a taxonomic ranking that is third in the hierarchy of biological classification after domain and kingdom.

hospital microbiome²⁰⁸ changed over the course of a new hospital opening.²⁰⁹ One should also note that in the Marotz study, low levels of SARS-CoV-2 were found on the environmental samples and the authors state that “the detected SARS-CoV-2 viral RNA was likely not in sufficient quantities to be infectious, consistent with previous findings of hospital surfaces.”²¹⁰

Summary of SARS-CoV-2’s Impact on Surfaces and Air

- 4.35 Microbes, including viruses, are normally found on surfaces (unless they have just been disinfected). A virus does not physically alter the surface on which it may come into contact. In contrast to the action of certain microbes such as molds that are able to penetrate and cause physical damage to some types of materials, there is no mechanism for viruses to do so. As previously discussed, a virus cannot replicate outside of a host cell, has no mechanism to move on its own or infiltrate a surface, and has no mechanism to physically or chemically alter the underlying surface.
- 4.36 With respect to viruses in the air, it is known that SARS-CoV-2, and other respiratory viruses, can remain infectious in aerosolized form within respiratory droplets for a limited period of time before settling on surfaces, although the natural rate of loss of infectivity is not established.²¹¹ Thus, it is possible for infectious viruses to be found in respiratory droplets in the air, particularly in the immediate vicinity of individuals infected with a

²⁰⁸ A microbiome is a “community of microorganisms (such as fungi, bacteria and viruses) that exists in a particular environment.” National Human Genome Research Institute (NHGRI). 2022. Microbiome. <https://www.genome.gov/genetics-glossary/Microbiome>. Accessed on 11/11/2022. Last updated on 5/10/2022. National Institutes of Health (NIH).

²⁰⁹ Lax, S., *et al.* 2017. Bacterial colonization and succession in a newly opened hospital. *Sci Transl Med* 9(391):eaah6500.

²¹⁰ Marotz, C., *et al.* 2021. SARS-CoV-2 detection status associates with bacterial community composition in patients and the hospital environment. *Microbiome* 9(1):132.

²¹¹ Samet, J. M., *et al.* 2021. SARS-CoV-2 indoor air transmission is a threat that can be addressed with science. *Proc Natl Acad Sci U S A* 118(45):e2116155118.

respiratory virus or immediately after they have left.^{212, 213, 214} However, the transient presence of viruses in the air²¹⁵ does not physically change the composition of the air that surrounds the respiratory droplets for several principal reasons. First, it is well-established that the presence of respiratory droplets in the air is limited due to their propensity to settle to upward facing horizontal surfaces.^{216, 217} Second, viruses do not alter the molecular composition of the air that surrounds the respiratory droplets, meaning for example that they do not deplete the oxygen, nor do they emit toxic fumes. Third, air is not static unless in a completely closed system. Generally, the volume of air in a room is constantly being mixed with air supplied from the heating, ventilation and air conditioning (HVAC) system or from adjacent rooms or from outdoor air which dilutes and removes aerosolized virus from the space. Fourth, viruses in respiratory droplets decay over a limited period of time rendering them non-infectious.

- 4.37 In general, laboratory studies have demonstrated that viruses can remain infectious only for a limited period of time generally ranging from minutes to hours when aerosolized and hours to days on surfaces, and the length of the time that a respiratory droplet retains its infectivity is a function of a combination of biological, physical, and chemical factors pertaining to the virus and environment. However, viruses in the absence of host cells are inert with respect to the environment.

²¹² Lednicky, J. A., *et al.* 2020. Viable SARS-CoV-2 in the air of a hospital room with COVID-19 patients. *Int J Infect Dis* 100:476-482.

²¹³ Rule, A. M., *et al.* 2018. Healthcare personnel exposure in an emergency department during influenza season. *PLoS One* 13(8):e0203223.

²¹⁴ Killingley, B., *et al.* 2016. The environmental deposition of influenza virus from patients infected with influenza A(H1N1)pdm09: Implications for infection prevention and control. *J Infect Public Health* 9(3):278-288.

²¹⁵ Here, air is defined as the gaseous substance surrounding the earth, which is a mixture mainly of oxygen and nitrogen. Air is composed of 78.08% Nitrogen, 20.95% Oxygen, 0.93% Argon, 0.04% CO₂, 0.002% Neon, and other trace gases. NASA. 2019. The Atmosphere: Getting a Handle on Carbon Dioxide. <https://climate.nasa.gov/news/2915/the-atmosphere-getting-a-handle-on-carbon-dioxide/#:~:text=What's%20in%20the%20Air%3F&text=By%20volume%2C%20the%20dry%20air,methane%2C%20nitrous%20oxide%20and%20ozone>. Accessed on 11/14/2022. Last updated on 10/09/2019.

²¹⁶ Atkinson, J., *et al.*, editors. 2009. Annex C: Respiratory Droplets in Natural Ventilation for Infection Control in Health-Care Settings. World Health Organization, Geneva.

²¹⁷ Rohit, A., *et al.* 2020. Fate of respiratory droplets in tropical vs temperate environments and implications for SARS-CoV-2 transmission. *Med Hypotheses* 144:109958.

G. How Surface Cleaning and Disinfection Disrupts Virus Structure

- 4.38 Dr. Carnethon suggests in her report that “adequate” surface cleaning was not feasible because “...respiratory droplets could be dispersed over a wide area and latch onto surfaces that could not be readily identified and cleaned. In addition, the scientific community did not know to what extent available cleaning products could eliminate the virus.”²¹⁸ As discussed in the previous section, viruses naturally degrade in the environment and scientific evidence shows that infectious virus has only been found to be absent or only in a small percentage of environmental samples collected in close proximity to COVID-19 patients (primarily in hospital settings). Thus, even without cleaning, scientific evidence suggests that only a very small fraction of the 24HF properties could have had infectious virus at any one time, such as in very close proximity to a person sick with COVID.
- 4.39 Dr. Carnethon’s statements further suggest that no information was available in March of 2020 regarding the effectiveness of different cleaning products. This is incorrect. As explained previously, SARS-CoV-2 was known very early on to be a coronavirus, and therefore, it was known that it was very likely to behave as other coronaviruses with respect to susceptibility to disinfectants and cleaning agents. As explained more below, this understanding is based on the pre-pandemic known structural properties of coronaviruses, e.g., enveloped viruses, and the understanding of how various disinfectants chemically disrupt and damage the structure of viruses.

Cleaners and Disinfectants Used at 24HF

- 4.40 After the review of the materials provided to me that were produced for this case, various cleaning agents were identified as potentially being used in 24HF facilities including Morning Mist, Virex II, Envirox H₂O Orange 2, and Biocharge.^{219, 220} The active ingredients in these products include quaternary amines, hydrogen peroxide, and

²¹⁸ Expert report of Mercedes R. Carnethon, Ph.D., October 21, 2022, Case No. 20-11558, p. 4-5.

²¹⁹ Deposition of Mr. Jeremy Gottlieb, dated June 17, 2022, Case No. 20-11558, Exhibit 3.

²²⁰ Deposition of Mr. Dan Larson, dated April 28, 2022, Case No. 20-11558, Exhibit 11.

detergents, which as explained below were known to be effective against enveloped viruses like SARS-CoV-2. It is unclear if additional cleaning agents were used and as such, the following sections will provide a general overview on the effectiveness and mechanisms of action of a variety of commonly used active ingredients on enveloped viruses, and in particular on SARS-CoV-2. Additionally, a portable hot water extractor was used.²²¹ As previously discussed, SARS-CoV-2 is sensitive to temperature and this type of hot water treatment may also contribute to increasing the viral decay rate if applied to contaminated materials.²²²

Mechanism of Action for Various Cleaning Chemistries

- 4.41 Cleaners with soaps or detergents have been recommended for regular cleaning to reduce the amount of pathogens and mitigate the risk of infection from surfaces.^{223, 224} In addition to the physical removal of viral particles during cleaning, surfactants in cleaners disrupt the viral membrane of an enveloped virus.^{225, 226, 227, 228, 229, 230} Other chemicals used in cleaning products disrupt the molecular and morphological structures as is briefly reviewed below and summarized in **Figure 11**.

²²¹ Deposition of Mr. Jeremy Gottlieb, dated June 17, 2022, Case No. 20-11558, Exhibit 3.

²²² Marchesi, I., *et al.* 2021. In vitro virucidal efficacy of a dry steam disinfection system against Human Coronavirus, Human Influenza Virus, and Echovirus. *J Occup Environ Hyg* 18(12):541-546.

²²³ CDC. 2021. Science Brief: SARS-CoV-2 and Surface (Fomite) Transmission for Indoor Community Environments. <https://www.cdc.gov/coronavirus/2019-ncov/more/science-and-research/surface-transmission.html>. Accessed on 11/11/2022. Last updated on 04/05/2021.

²²⁴ Artasensi, A., *et al.* 2021. Back to Basics: Choosing the Appropriate Surface Disinfectant. *Antibiotics*.

²²⁵ Gerlach, M., *et al.* 2020. Rapid SARS-CoV-2 inactivation by commonly available chemicals on inanimate surfaces. *J Hosp Infect* 106(3):633-634.

²²⁶ Dehbandi, R., and M. A. Zazouli. 2020. Stability of SARS-CoV-2 in different environmental conditions. *Lancet Microbe* 1(4):e145.

²²⁷ Jahromi, R., *et al.* 2020. Synergistic effects of anionic surfactants on coronavirus (SARS-CoV-2) virucidal efficiency of sanitizing fluids to fight COVID-19. *Food Chem Toxicol* 145:111702.

²²⁸ Matsumoto, T. 2020. Report on Efficacy Assessment of Disinfecting Substances Alternative to Alcohol Against SARS-CoV-2 - Executive Summary -. <https://www.nite.go.jp/data/000115862.pdf>. Accessed on 05/10/2022. Last updated on June 2020. National Institute of Technology and Evaluation.

²²⁹ Almeida, C. F., *et al.* 2022. The Efficacy of Common Household Cleaning Agents for SARS-CoV-2 Infection Control. *Viruses* 14(4).

²³⁰ Owen, L., *et al.* 2021. The Stability of Model Human Coronaviruses on Textiles in the Environment and during Health Care Laundering. *mSphere* 6(2).

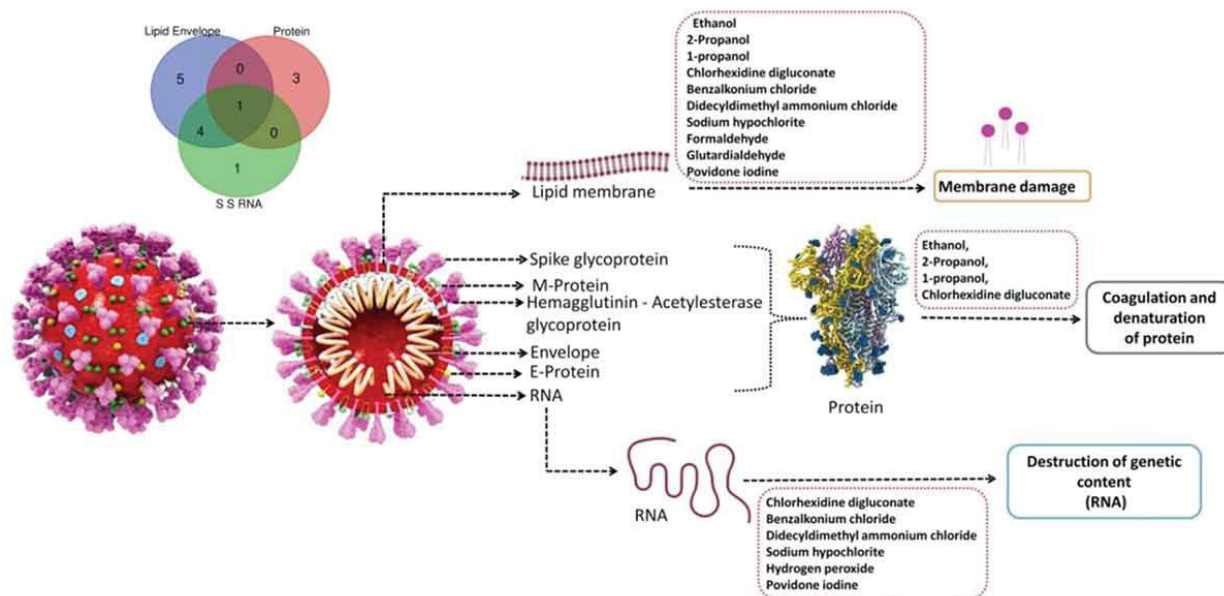


Figure 11. Mechanism of disinfection of coronaviruses by various types of disinfectants. Reproduced from Khokhar *et al.*²³¹

4.42 Hypochlorite-based solutions (e.g., bleach) and other compounds that release free available chlorine have a broad spectrum of antimicrobial activity. Hypochlorous acid (HOCl) is thought to be the major contributor to the microbicidal activity of hypochlorite-based disinfectants.²³² For instance, low concentrations of hypochlorite (0.05%) have been shown to be effective against rotaviruses, while higher concentrations (0.5%) are required for highly resistant human pathogens such as the bacteria *Clostridium difficile* or the fungus *Candida auris*.²³³ The viral inactivation can result from different factors such as protein synthesis inhibition, DNA damage or impaired DNA synthesis. This disinfectant has been

²³¹ Khokhar, M., *et al.* 2020. Viricidal treatments for prevention of coronavirus infection. *Pathog Glob Health* 114(7):349-359.

²³² Al-Sayah, M. H. 2020. Chemical disinfectants of COVID-19: an overview. *J Water Health* 18(5):843-848.

²³³ WHO. 2020. Cleaning and disinfection of environmental surfaces in the context of COVID-19. <https://www.who.int/publications/i/item/cleaning-and-disinfection-of-environmental-surfaces-in-the-context-of-covid-19>. Accessed on 11/12/2022. Last updated on 05/16/2020.

shown to be effective against several viruses in 10 minutes with concentrations as low as 200 ppm of available chlorine.²³⁴

- 4.43 Alcohol at concentrations between 60-90% is a virucidal agent. Ethanol is effective against a broad range of viruses. Isopropyl alcohol has also shown to be active against enveloped viruses.^{235, 236, 237} The virucidal action of alcohols is thought to be protein denaturation.²³⁸
- 4.44 Hydrogen peroxide is an active agent against a wide range of microorganisms including viruses, bacteria and yeasts. It acts by producing hydroxyl free radicals that degrade various cellular components including membrane lipids and nucleic acids.^{239, 240}
- 4.45 Quaternary ammonium compounds have shown to be virucidal against enveloped viruses through enzyme inactivation, protein denaturation and cell membrane disruption.^{241, 242, 243} However, their activity against hydrophilic (non-enveloped) viruses, spores, mycobacterium and gram-negative bacteria is limited.^{244, 245, 246}

²³⁴ CDC. 2019. Guideline for Disinfection and Sterilization in Healthcare Facilities, 2008; Update: May 2019. <https://www.cdc.gov/infectioncontrol/pdf/guidelines/disinfection-guidelines-H.pdf>. Accessed on 11/11/2022. Last updated on 5/1/2019.

²³⁵ Golin, A. P., *et al.* 2020. Hand sanitizers: A review of ingredients, mechanisms of action, modes of delivery, and efficacy against coronaviruses. *Am J Infect Control* 48(9):1062-1067.

²³⁶ Khokhar, M., *et al.* 2020. Viricidal treatments for prevention of coronavirus infection. *Pathog Glob Health* 114(7):349-359.

²³⁷ McDonnell, G., and A. D. Russell. 1999. Antiseptics and disinfectants: activity, action, and resistance. *Clin Microbiol Rev* 12(1):147-179.

²³⁸ Khokhar, M., *et al.* 2020. Viricidal treatments for prevention of coronavirus infection. *Pathog Glob Health* 114(7):349-359.

²³⁹ Khokhar, M., *et al.* 2020. Viricidal treatments for prevention of coronavirus infection. *Pathog Glob Health* 114(7):349-359.

²⁴⁰ McDonnell, G., and A. D. Russell. 1999. Antiseptics and disinfectants: activity, action, and resistance. *Clin Microbiol Rev* 12(1):147-179.

²⁴¹ Khokhar, M., *et al.* 2020. Viricidal treatments for prevention of coronavirus infection. *Pathog Glob Health* 114(7):349-359.

²⁴² Maillard, J.-Y., *et al.* 2013. Virucidal Activity of Microbicides. pp. 178-207. In: Russell, Hugo & Ayliffe's.

²⁴³ McDonnell, G., and A. D. Russell. 1999. Antiseptics and disinfectants: activity, action, and resistance. *Clin Microbiol Rev* 12(1):147-179.

²⁴⁴ Khokhar, M., *et al.* 2020. Viricidal treatments for prevention of coronavirus infection. *Pathog Glob Health* 114(7):349-359.

²⁴⁵ Maillard, J.-Y., *et al.* 2013. Virucidal Activity of Microbicides. Pp. 178-207. In: Russell, Hugo & Ayliffe's.

²⁴⁶ McDonnell, G., and A. D. Russell. 1999. Antiseptics and disinfectants: activity, action, and resistance. *Clin Microbiol Rev* 12(1):147-179.

- 4.46 Other non-chemical mechanisms used as disinfection methods such as heat or UV are also effective against viruses. Studies have shown that exposure to UV radiation results in both protein and genetic material damage, while heat treatments induce protein conformational changes that decrease the virion ability to bind to host cells.^{247, 248}
- 4.47 Once inactivated, viral particles are not able to use the host machinery to replicate even if exposed to host cells. Upon exposure to the chemical or physical disinfection methods, the structure of the viral particles is disrupted (e.g., protein denaturation, capsid destabilization, envelope disruption, and/or DNA damage). Afterwards, only the inert viral components (lipids, proteins and nucleic acids) will temporarily remain on the disinfected surface and will eventually degrade into even smaller molecules and be covered by micro-sized particles composing dust. Because proteins and nucleic acids are also the building blocks of microbes, plants, and animals, the same types of inert materials are likewise left behind on surfaces after a disinfection process in the absence of SARS-CoV-2, for example from other microbes or components of human origin, such as skin debris. Thus, appropriately disinfected surfaces do not transmit SARS-CoV-2.

Surface Disinfection from SARS-CoV-2

- 4.48 As mentioned under “Introduction to SARS-CoV-2” above, the structural properties of viruses will dictate the viral susceptibility to specific disinfectants. The structure of SARS-CoV-2 includes a fragile outer lipid envelope, which makes it more vulnerable to common disinfectants as compared to other non-enveloped viruses.^{249, 250, 251, 252} For reference, the hierarchy of susceptibility of human pathogenic viruses to chemical disinfectants is

²⁴⁷ Wigginton, K. R., *et al.* 2012. Virus Inactivation Mechanisms: Impact of Disinfectants on Virus Function and Structural Integrity. *Environmental Science & Technology* 46(21):12069-12078.

²⁴⁸ Gidari, A., *et al.* 2021. SARS-CoV-2 Survival on Surfaces and the Effect of UV-C Light. *Viruses* 13(3).

²⁴⁹ Chin, A. W. H., *et al.* 2020. Stability of SARS-CoV-2 in different environmental conditions. *Lancet Microbe* 1(1):e10.

²⁵⁰ Cimolai, N. 2020. Environmental and decontamination issues for human coronaviruses and their potential surrogates. *J Med Virol* 92(11):2498-2510.

²⁵¹ Ijaz, M. K., *et al.* 2020. Microbicidal actives with virucidal efficacy against SARS-CoV-2. *Am J Infect Control* 48(8):972-973.

²⁵² Ijaz, M. K., *et al.* 2021. Microbicidal actives with virucidal efficacy against SARS-CoV-2 and other beta- and alpha-coronaviruses and implications for future emerging coronaviruses and other enveloped viruses. *Sci Rep* 11(1):5626.

illustrated in **Figure 12**. Enveloped viruses exhibit the highest vulnerability to chemical disinfectants, followed by large non-enveloped viruses, and finally small non-enveloped viruses. Even at the beginning of the pandemic, SARS-CoV-2 was expected to be as susceptible to biocides as other common human endemic viruses such as influenza (flu), which are found in the same category.

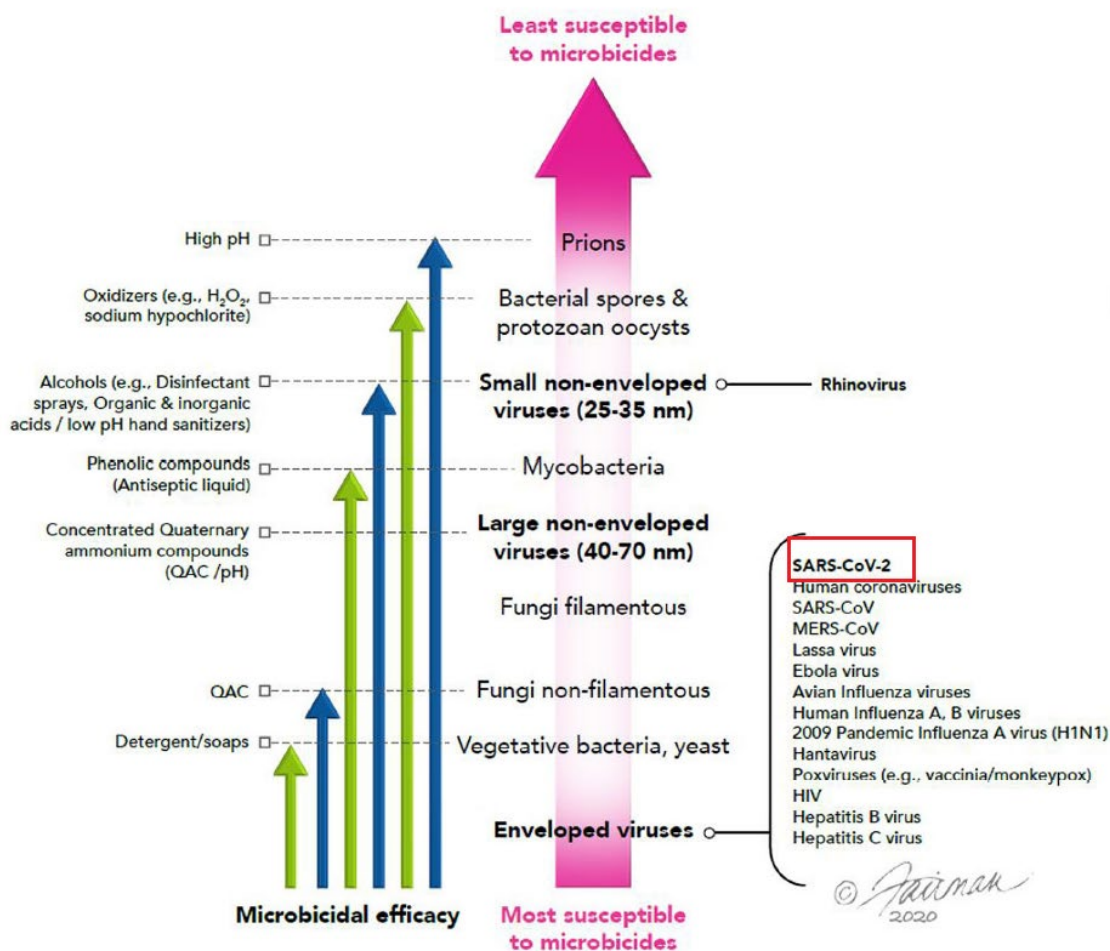


Figure 12. Hierarchy of susceptibility of human pathogens to chemical disinfectants. This figure includes other infectious pathogens for comparison. SARS-CoV-2 is boxed in red. Reproduced from Ijaz *et al.*²⁵³

²⁵³ Ijaz, M. K., *et al.* 2021. Microbicidal actives with virucidal efficacy against SARS-CoV-2 and other beta- and alpha-coronaviruses and implications for future emerging coronaviruses and other enveloped viruses. *Sci Rep* 11(1):5626.

4.48 Published studies with SARS-CoV-2 have confirmed its susceptibility to common biocides.^{254, 255, 256, 257, 258, 259, 260, 261} For example, Chin *et al.* found SARS-CoV-2 was susceptible to bleach, hand soap, ethanol, povidone-iodine, chloroxylenol, chlorohexidine, and benzalkonium chloride.²⁶² In other work, Ijaz *et al.* confirmed the efficacy of antiseptic liquid, hand sanitizer gel, liquid hand wash, bar soap, surface cleanser, disinfectant wipes, and disinfectant spray using standardized methods.²⁶³ Additionally, a study by the Japanese National Institute of Technology and Evaluation found nine surfactants to be effective against SARS-CoV-2, including sodium linear alkylbenzene sulfonates, alkyl glycosides, alkyldimethylamine oxide, benzalkonium chloride, benzethonium chloride, dialkyldimethylammonium chloride, polyoxyethylene alkyl ether, potassium soap, and sodium soap.²⁶⁴ Many types of disinfectants that have been successfully used with other coronaviruses have been shown to be effective against SARS-CoV-2, and therefore it is expected that use of a recommended disinfectant on a surface following the manufacturer's guidelines does not impact its functionality. To this end, the EPA maintains a list ("List N") of products for disinfection from SARS-CoV-2,

²⁵⁴ Chin, A. W. H., *et al.* 2020. Stability of SARS-CoV-2 in different environmental conditions. *Lancet Microbe* 1(1):e10.

²⁵⁵ Cimolai, N. 2020. Environmental and decontamination issues for human coronaviruses and their potential surrogates. *J Med Virol* 92(11):2498-2510.

²⁵⁶ Ijaz, M. K., *et al.* 2020. Microbicidal actives with virucidal efficacy against SARS-CoV-2. *Am J Infect Control* 48(8):972-973.

²⁵⁷ Fischer, R., *et al.* 2020. Effectiveness of N95 Respirator Decontamination and Reuse against SARS-CoV-2 Virus. *Emerging Infectious Disease journal* 26(9):2253.

²⁵⁸ Kratzel, A., *et al.* 2020. Inactivation of Severe Acute Respiratory Syndrome Coronavirus 2 by WHO-Recommended Hand Rub Formulations and Alcohols. *Emerging Infectious Disease journal* 26(7):1592.

²⁵⁹ Ogilvie, B. H., *et al.* 2021. Alcohol-free hand sanitizer and other quaternary ammonium disinfectants quickly and effectively inactivate SARS-CoV-2. *J Hosp Infect* 108:142-145.

²⁶⁰ Almeida, C. F., *et al.* 2022. The Efficacy of Common Household Cleaning Agents for SARS-CoV-2 Infection Control. *Viruses* 14(4).

²⁶¹ Lee, G. H., *et al.* 2022. Comparative efficacy evaluation of disinfectants against severe acute respiratory syndrome coronavirus-2. *J Hosp Infect* 131:12-22.

²⁶² Chin, A. W. H., *et al.* 2020. Stability of SARS-CoV-2 in different environmental conditions. *Lancet Microbe* 1(1):e10. Supplementary information.

²⁶³ Ijaz, M. K., *et al.* 2020. Microbicidal actives with virucidal efficacy against SARS-CoV-2. *Am J Infect Control* 48(8):972-973.

²⁶⁴ Matsumoto, T. 2020. Report on Efficacy Assessment of Disinfecting Substances Alternative to Alcohol Against SARS-CoV-2 - Executive Summary -. <https://www.nite.go.jp/data/000115862.pdf>. Accessed on 05/10/2022. Last updated on June 2020. National Institute of Technology and Evaluation.

which contains 629 products as of November 12, 2022.²⁶⁵ Other non-chemical methods, such as UV-disinfection^{266, 267} and heat,²⁶⁸ have also been shown to be effective against SARS-CoV-2.

²⁶⁵ Environmental Protection Agency (EPA). 2022. List N Tool: COVID-19 Disinfectants. <https://cfpub.epa.gov/wizards/disinfectants/>. Accessed on 11/11/2022.

²⁶⁶ Food and Drug Administration (FDA). 2021. UV Lights and Lamps: Ultraviolet-C Radiation, Disinfection, and Coronavirus. <https://www.fda.gov/medical-devices/coronavirus-covid-19-and-medical-devices/uv-lights-and-lamps-ultraviolet-c-radiation-disinfection-and-coronavirus>. Accessed on 11/11/2022. Last updated on 02/01/2021.

²⁶⁷ Tomas, A. L., *et al.* 2022. UV-C irradiation-based inactivation of SARS-CoV-2 in contaminated porous and non-porous surfaces. *J Photochem Photobiol B* 234:112531.

²⁶⁸ Abraham, J. P., *et al.* 2020. Using heat to kill SARS-CoV-2. *Rev Med Virol* 30(5):e2115.

5. Summary of Opinions

- 5.1 In this report, technical information relating to the science of how viruses interact with surfaces, of viral persistence in the environment, and of disinfection methods is described. The report describes general information about viruses, as well as specific information about coronaviruses and SARS-CoV-2 as far as it is publicly available in the scientific literature at the time of the writing of this report. Based on the information available to me as well as my education, training and experience, I offer the following opinions, as well as those expressed in the body of this report, to a reasonable degree of scientific certainty.
- 5.2 Overall, it is my opinion that there is no scientific basis for the assertion that SARS-CoV-2 adversely affects the surfaces or surrounding air it contacts, or that this coronavirus remains infectious after either general degradation or disinfection by one of a wide range of effective means. In other words, SARS-CoV-2 does not physically alter, change, or damage the air that surrounds respiratory droplets that contain infectious virus or the surface on which these droplets settle, thereby allowing for easy disinfection of the surface. My opinion is based on the information outlined in my report and summarized in the sub-bullets below:
- (1) SARS-CoV-2 generally behaves similarly to other coronaviruses with respect to its physical, chemical, and biological interaction with inanimate surfaces.
 - (2) SARS-CoV-2, like other viruses, requires specific interactions with animate host cells that express particular receptors to replicate. SARS-CoV-2, as with other viruses, cannot replicate in the environment apart from their host cells.
 - (3) Outside of the interaction with its host cells, SARS-CoV-2, like other viruses, can only adsorb to surfaces and has no mechanism to infiltrate the surface, change, impact, or damage a surface's chemical or physical composition.
 - (4) SARS-CoV-2 cannot physically alter or damage the surface on which it adsorbs.
 - (5) SARS-CoV-2 can be aerosolized in respiratory droplets and be airborne for a limited period of time in a manner similar to other respiratory viruses but cannot physically alter the chemical composition of the surrounding air.

- (6) SARS-CoV-2, like other viruses, can persist in the environment away from host cells for a limited period of time, but the length of the time that it retains its infectivity is a function of a combination of biological, physical, and chemical factors pertaining to the virus and environment. In relation to this:
 - (a) Laboratory studies have shown that SARS-CoV-2 can retain infectivity on the order of hours to days on surfaces and on the order of minutes to hours in aerosols, although these studies may overestimate the persistence time in the real-world.
 - (b) Real-world measurements suggest that the persistence of SARS-CoV-2 in the environment is more limited, as infectious virus was infrequently detected when samples were specifically tested for it.
- (7) Surfaces with SARS-CoV-2 on them, like other viruses, can be disinfected using standard disinfection methods to hasten the natural viral degradation process. The effectiveness of a wide range of disinfectants has been studied and shown to be effective against SARS-CoV-2.
- (8) Appropriately disinfected surfaces do not transmit SARS-CoV-2. Since a wide range of disinfectant chemistries and non-chemical methods (e.g., UV-disinfection, heat) have been shown to be effective against SARS-CoV-2, there is no basis to conclude that the ability to effectively disinfect surfaces is limited in a practical way.

Appendix A: Dr. Sauer-Budge's CV



Exponent®
Engineering & Scientific Consulting

Alexis Sauer-Budge, Ph.D.

Senior Managing Scientist | Biomedical Engineering & Sciences
1075 Worcester St. | Natick, MA 01760
(508) 652-8563 tel | asauerbudge@exponent.com

Professional Profile

Dr. Sauer-Budge specializes in the intersection between biology and materials. Trained in chemistry and biophysics, she applies her interdisciplinary experience to the development and design analysis of products for the medical device, biotechnology, and pharmaceutical industries. She has particular expertise in clinical diagnostics (the testing and identification of biological materials), including nucleic acids (DNA and RNA), proteins, microbes (bacteria, fungi, antimicrobial resistance), and human specimens (blood, urine, saliva, etc.). Throughout her career, Dr. Sauer-Budge has been involved in the research and development of biosensors, sample preparation techniques, microfluidic devices, anti-fouling coatings, wearable devices for the continuous monitoring of biomarkers, implants, 3D bioprinting, bench-top instrumentation, and multi-functional surgical tools.

Dr. Sauer-Budge assists clients in device and in vitro diagnostics design, identification of microbial, biological, and chemical contaminants, assay development (immunoassays, nucleic acid tests, cell-based assays), testing of the impact microbial growth on materials, biocompatible/non-fouling coatings, and scale-up for manufacturing. Dr. Sauer-Budge works with companies at all stages as well as in the government and legal sectors. She is an active participant in peer review, including serving as a standing member of an NIH review committee and has published more than 35 articles.

Prior to joining Exponent, Dr. Sauer-Budge led the biomedical/biotechnology group at the Fraunhofer Center for Manufacturing Innovation at Boston University for 10 years. She worked in the space between academia and industry, conducting applied research in the areas of medical diagnostics, devices, and instrumentation. She managed translational programs, helping to transition technologies from the bench through scale-up and FDA readiness. Prior to Fraunhofer, Dr. Sauer-Budge worked at BioScale, a start-up focused on commercializing a bioMEMS resonating membrane platform technology for the clinical diagnostic and food safety markets. Her graduate work was in the laboratory of Prof. Daniel Branton developing single molecule sequencing technologies that is now commercialized by Oxford Nanopore.

Academic Credentials & Professional Honors

Ph.D., Biophysics, Harvard University, 2002

M.S., Chemistry, Stanford University, 1997

B.S., Chemistry, Stanford University, 1996

Academic Appointments

Adjunct Associate Professor, Biomedical Engineering, Boston University, 2017-2018

Adjunct Research Assistant Professor, Biomedical Engineering, Boston University, 2009-2017

Prior Experience

Senior Research Scientist, Fraunhofer Center for Manufacturing Innovation, 2007-2017

Director of Microbial Assay Development, BioScale, Inc., 2005-2007

Senior Scientist, BioScale, Inc, 2003-2005

Consultant, Eagle Research and Development, 2000-2003

Research Assistant, Hauser Chemical Research, 1993-1995

Professional Affiliations

American Association for Clinical Chemistry (AACC)

Northeast Branch - American Society for Microbiology (ASM)

Patents

US Patent Application #16/486,114 "Pipetting Devices and Methods of Using the Same." July 2020. A. Sauer-Budge, H. Wirz, S.J. Brookfield, N. Pollock, R. Janzen.

US Patent # 9199250 B2 "Disposable Separator/Concentrator Device and Method of Use." December 2015. A. Sauer-Budge, A. Size, H. Wirz, A. Sharon.

US Patent #9046483 B2 "Characterization of hybridized polymer molecules based on monomer-interface interactions." July 2015. T. Denison, A. Sauer-Budge, J. Golovchenko, A. Meller, E. Brandin, D. Branton.

US Patent #8986528 B2 "Characterization of hybridized polymer molecules based on monomer-interface interactions." March 2015. T. Denison, A. Sauer-Budge, J. Golovchenko, A. Meller, E. Brandin, D. Branton.

US Patent # 8,785,148 "Method and Device for Rapid Detection of Bacterial Antibiotic Resistance/Susceptibility" July 2014. A. Sauer-Budge, A. Sharon, M. Kalashnikov, H. Wirz.

US Patent # 8,227,261, "Methods and apparatus for assay measurements." July 2012. B. Masters, M. Miller, A. Sauer-Budge.

US Patent # 7,632,638 B2 "Methods and apparatus for detecting viruses using an acoustic device." December 2009. A. Sauer-Budge, B. Masters, M. Miller, M. Lundstrom.

US Patent # 7,629,137 B2 "Methods and apparatus for detecting bacteria using an acoustic device." December 2009. A. Sauer-Budge, E. Fitch, B. Masters, M. Miller, M. Lundstrom.

US Patent # 6,673,615 "Characterization of hybridized polymer molecules based on monomer-interface interactions." January 2004. T. Denison, A. Sauer, J. Golovchenko, A. Meller, E. Brandin, D. Branton.

US Patent # 6,362,002 B1 "Characterization of hybridized polymer molecules based on monomer-interface interactions." March 2002. T. Denison, A. Sauer, J. Golovchenko, A. Meller, E. Brandin, D. Branton.

Publications

Spink, S., F. Teng, V. Pera, H. Peterson, T. Cormier, A. Sauer-Budge, D. Chargin, S. Brookfield, A. Eggebrecht, N. Y. Ko, and D. Roblyer. 2021. High optode-density wearable diffuse optical probe for monitoring paced breathing hemodynamics in breast tissue. *J Biomed Opt* 26(6).

Wirz, H., S. Teufelhart, C. McBeth, R. Gyurko, S. Dibart, and A. Sauer-Budge. 2020. Design and ex vivo characterization of narrow implants with custom piezo-activated osteotomy for patients with substantial bone loss. *Clinical and Experimental Dental Research*. 6(3): 336-344.

Sauer-Budge, A. F. 2019. New technologies for the rapid identification of drug-resistant bacteria. *TechConnect Briefs*:310-313.

Gladman, A. S., M. Garcia-Leiner, and A. F. Sauer-Budge. 2019. Emerging polymeric materials in additive manufacturing for use in biomedical applications. 6(1):1-20.

Gutermuth, A., J. Maassen, E. Harnisch, D. Kuhlen, A. Sauer-Budge, C. Skazik-Voogt, and K. Engelmann. 2019. Descemet's Membrane Biomimetic Microtopography Differentiates Human Mesenchymal Stem Cells Into Corneal Endothelial-Like Cells. *Cornea* 38(1):110-119.

Fernandez-Carballo, B. L., C. McBeth, I. McGuinness, M. Kalashnikov, C. Baum, S. Borros, A. Sharon, and A. F. Sauer-Budge. 2018. Continuous-flow, microfluidic, qRT-PCR system for

RNA virus detection. *Anal Bioanal Chem* 410(1):33-43.

Ganser, P., C. Baum, D. Chargin, A. F. Sauer-Budge, and A. Sharon. 2018. A practical approach for the optimization of channel integrity in the sealing of shallow microfluidic devices made from cyclic olefin polymer. *Biomed Microdevices* 20(2):24.

Kalashnikov, M., J. C. Lee, and A. F. Sauer-Budge. 2018. Optimization of Stress-Based Microfluidic Testing for Methicillin Resistance in *Staphylococcus aureus* Strains. *Diagnostics (Basel)* 8(2).

Sauer-Budge, A. F., S. J. Brookfield, R. Janzen, S. McGray, A. Boardman, H. Wirz, and N. R. Pollock. 2017. A novel device for collecting and dispensing fingerstick blood for point of care testing. *PLoS One* 12(8):e0183625.

Kalashnikov, M., M. Mueller, C. McBeth, J. C. Lee, J. Campbell, A. Sharon, and A. F. Sauer-Budge. 2017. Rapid phenotypic stress-based microfluidic antibiotic susceptibility testing of Gram-negative clinical isolates. *Sci Rep* 7(1):8031.

McBeth, C., A. Gutermuth, J. Ochs, A. Sharon, and A. F. Sauer-Budge. 2017. Automated Tissue Dissociation for Rapid Extraction of Viable Cells. *Procedia CIRP* 65:88-92.

Kulik, M., J. Ochs, N. König, C. McBeth, A. Sauer-Budge, A. Sharon, and R. Schmitt. 2017. Parallelization in Automated Stem Cell Culture. *Procedia CIRP* 65:242-247.

McBeth, C., J. Lauer, M. Ottersbach, J. Campbell, A. Sharon, and A. F. Sauer-Budge. 2017. 3D bioprinting of GelMA scaffolds triggers mineral deposition by primary human osteoblasts. *Biofabrication* 9(1):015009.

Teng, F., T. Cormier, A. Sauer-Budge, R. Chaudhury, V. Pera, R. Istfan, D. Chargin, S. Brookfield, N. Y. Ko, and D. M. Roblyer. 2017. Wearable near-infrared optical probe for continuous monitoring during breast cancer neoadjuvant chemotherapy infusions. *J Biomed Opt* 22(1):14001.

Campbell, J., C. McBeth, M. Kalashnikov, A. K. Boardman, A. Sharon, and A. F. Sauer-Budge. 2016. Microfluidic advances in phenotypic antibiotic susceptibility testing. *Biomed Microdevices* 18(6):103.

Boardman, A. K., W. S. Wong, W. R. Premasiri, L. D. Ziegler, J. C. Lee, M. Miljkovic, C. M. Klapperich, A. Sharon, and A. F. Sauer-Budge. 2016. Rapid Detection of Bacteria from Blood with Surface-Enhanced Raman Spectroscopy. *Anal Chem* 88(16):8026-8035.

Premasiri, W. R., J. C. Lee, A. Sauer-Budge, R. Theberge, C. E. Costello, and L. D. Ziegler. 2016. The biochemical origins of the surface-enhanced Raman spectra of bacteria: a metabolomics profiling by SERS. *Anal Bioanal Chem* 408(17):4631-4647.

Fernandez-Carballo, B. L., I. McGuinness, C. McBeth, M. Kalashnikov, S. Borros, A. Sharon, and

A. F. Sauer-Budge. 2016. Low-cost, real-time, continuous flow PCR system for pathogen detection. *Biomed Microdevices* 18(2):34.

Keenan, M., C. Howard, T. Tate, I. McGuinness, A. Sauer-Budge, J. Black, U. Utzinger, and J. K. Barton. 2016. Design of an everting balloon to deploy a microendoscope to the fallopian tubes. *SPIE BiOS: Photonic Therapeutics and Diagnostics XII*. SPIE.

Teng, F., T. Cormier, A. Sauer-Budge, and D. M. Roblyer. 2016. A wearable optical device for continuous monitoring during neoadjuvant chemotherapy infusions. *SPIE BiOS: Optical Diagnostics and Sensing XVI: Toward Point-of-Care Diagnostics*. SPIE.

Campbell, J., N. Pollock, A. Sharon, and A. F. Sauer-Budge. 2015. Development of an automated on-chip bead-based ELISA platform. *Anal Methods* 7(19):8472-8477.

Campbell, J., I. McGuinness, H. Wirz, A. Sharon, and A. F. Sauer-Budge. 2015. Multimaterial and Multiscale Three-Dimensional Bioprinter. *Journal of Nanotechnology in Engineering and Medicine* 6(2):021005-021007.

Boardman, A. K., J. Campbell, H. Wirz, A. Sharon, and A. F. Sauer-Budge. 2015. Rapid microbial sample preparation from blood using a novel concentration device. *PLoS One* 10(2):e0116837.

Rosen, J. E., A. Size, Y. Yang, A. Sharon, and A. Sauer-Budge. 2015. Artificial hand for minimally invasive surgery: design and testing of initial prototype. *Surg Endosc* 29(1):61-67.

Briggs, J. C., O. A'amar, I. Bigio, J. E. Rosen, S. L. Lee, A. Sharon, and A. F. Sauer-Budge. 2014. Integrated Device for in Vivo Fine Needle Aspiration Biopsy and Elastic Scattering Spectroscopy in Preoperative Thyroid Nodules. *Journal of Medical Devices* 8(2):021003-021006.

Kalashnikov, M., J. Campbell, J. C. Lee, A. Sharon, and A. F. Sauer-Budge. 2014. Stress-induced antibiotic susceptibility testing on a chip. *J Vis Exp* (83):e50828.

Byrnes, S., A. Fan, J. Trueb, F. Jareczek, M. Mazzochette, A. Sharon, A. F. Sauer-Budge, and C. M. Klapperich. 2013. A Portable, Pressure Driven, Room Temperature Nucleic Acid Extraction and Storage System for Point of Care Molecular Diagnostics. *Anal Methods* 5(13):3177-3184.

Boardman, A. K., S. Allison, A. Sharon, and A. F. Sauer-Budge. 2013. Comparison of anti-fouling surface coatings for applications in bacteremia diagnostics. *Anal Methods* 5(1):273-280.

Sauer-Budge, A. F., A. K. Boardman, S. Allison, H. Wirz, D. Foss, and A. Sharon. 2013. Materials and Surface Properties Optimization to Prevent Biofouling of a Novel Bacterial Concentrator. *Procedia CIRP* 5:185-188.

Mirsky, P., A. Chatterjee, A. F. Sauer-Budge, and A. Sharon. 2012. An automated, parallel processing approach to biomolecular sample preparation. *J Lab Autom* 17(2):116-124.

Kalashnikov, M., J. C. Lee, J. Campbell, A. Sharon, and A. F. Sauer-Budge. 2012. A microfluidic platform for rapid, stress-induced antibiotic susceptibility testing of *Staphylococcus aureus*. *Lab Chip* 12(21):4523-4532.

Wirz, H., A. F. Sauer-Budge, J. Briggs, A. Sharpe, S. Shu, and A. Sharon. 2012. Automated production of plant-based vaccines and pharmaceuticals. *J Lab Autom* 17(6):449-457.

Premasiri, W. R., A. F. Sauer-Budge, J. C. Lee, C. M. Klapperich, and L. D. Ziegler. 2012. Rapid bacterial diagnostics via surface enhanced Raman microscopy. *Spectroscopy (Springf)* 27(6):s8-31.

Campbell, J., P. Mirsky, A. Chatterjee, A. Sharon, and A. F. Sauer-Budge. 2012. A semi-automated liquid handler for parallel sample preparation. *Biotech International (May / June)*:23-25.

Gruentzig, A. W., C. M. Klapperich, A. Sharon, J. Braman, A. Chatterjee, and A. F. Sauer-Budge. 2011. A new DNA extraction method for automated food analysis. *Analytical Methods* 3:1507-1513.

Sauer-Budge, A. F., and A. Sharon. 2011. Editorial for the special issue of RCIM on translational research — Where engineering meets medicine. *Robotics and Computer-Integrated Manufacturing* 27(2):235-236.

Size, A., A. Sharon, and A. Sauer-Budge. 2011. An automated low cost instrument for simultaneous multi-sample tissue homogenization. *Robotics and Computer-Integrated Manufacturing* 27(2):276-281.

Sauer-Budge, A.F., P. Mirer, A. Chatterjee, N. Pollock, C. Klapperich, and A. Sharon. Integrated lab-on-a-chip Influenza diagnostic designed for low cost manufacturing. *Techworld 2010*, 2010. Anaheim, CA.

Chatterjee, A., P. L. Mirer, E. Zaldivar Santamaria, C. Klapperich, A. Sharon, and A. F. Sauer-Budge. 2010. RNA isolation from mammalian cells using porous polymer monoliths: an approach for high-throughput automation. *Anal Chem* 82(11):4344-4356.

Sauer-Budge, A. F., P. Mirer, A. Chatterjee, C. M. Klapperich, D. Chargin, and A. Sharon. 2009. Low cost and manufacturable complete microTAS for detecting bacteria. *Lab Chip* 9(19):2803-2810.

Sauer-Budge, A. F., J. A. Nyamwanda, D. K. Lubensky, and D. Branton. 2003. Unzipping kinetics of double-stranded DNA in a nanopore. *Phys Rev Lett* 90(23):238101.

Sauer-Budge, A. and Branton, D. (2002). "Unzipping Double-Stranded DNA Molecule by Molecule through a Nanopore." Thesis presented to Harvard University in partial fulfillment of the requirements for the degree of Doctor of Philosophy in Biophysics. October.

Sauer, A. and Zare, R. (1997). "The Interaction Of Peptide Nucleic Acid With Complementary Single-Stranded DNA And Double-Stranded DNA In The Development Of A Genetic Screening Protocol" Thesis presented to Stanford University in partial fulfillment of the requirements for the degree of Master of Science in Chemistry. June.

Presentations

Sauer-Budge, A. (2021) Panel speaker "Wonder Women (and Men) – Lessons Learned in the Pandemic Trenches" DRI Women in the Law Seminar.

Sauer-Budge, A., Bergerson, C. (2021) Webinar. "The Latest in COVID-19 Testing: Where We Are and Where We Are Going."

Khanolkar, A., Walder, T., Sauer-Budge, A. (2020) Panel speaker "Material Innovations Driving Innovation & Quality Manufacturing" Conference Track: Product Development Innovation, BIOMEDigital 2020.

Sauer-Budge, A., Bergerson, C., and Lipp, J. (2020) Webinar. "COVID-19 Diagnostic Tests: Perspectives from the Scientific, Regulatory, and Legal Communities."

Sauer-Budge, A. (2020) Panel speaker "Product Liability and the Coronavirus." American Bar Association (ABA) Women in Products Liability Virtual Regional CLE Program.

Sauer-Budge, A. (2019) Track Chair. "Panel: How Miniaturization is Driving Design in Top Medical Device Categories" BIOMEDevice Boston, Track A: Product Development, Boston, MA.

Sauer-Budge, A. (2017) Invited talk. "Rapid Sample to Answer Antibiotic Susceptibility Testing for Bacteremia" Lab-on-a-Chip & Microfluidics World Congress 2017, San Diego, CA.

Sauer-Budge, A. (2017) Invited talk. "Automated tissue dissociation for rapid extraction of viable cells" CIRP Biomanufacturing, Chicago, IL.

Kalashnikov M, Mueller M, McBeth C, Lee JC, Sharon A, Sauer-Budge, A. (2017) Poster "Phenotypic stress-based antibiotic susceptibility testing of Gram-negative clinical isolates" Gordon Research Conference, Microfluidics, Italy.

Sauer-Budge, A. (2017) Invited talk. "Assay development while designing for manufacturing" Microfluidics 8.4, Boston, MA.

Sauer-Budge, A. (2017) Invited talk. "Plasma surface modifications for microfluidic and diagnostic devices" PlasmaTreat Open House.

Sauer-Budge, A. (2017) Invited talk. "Empowering Rapid Diagnostics with Sample Preparation Methodologies" Sample Prep, Washington DC.

Sauer-Budge, A. (2016) Invited Talk. "Rapid phenotypic methods for diagnosing infections and antibiotic susceptibility testing" BioDefense Global Summit, Baltimore, MD.

Sauer-Budge, A. (2015) Invited Talk. "Isolation of Dilute Pathogens from Blood" Select Bio Circulating Biomarkers World Congress 2015, Boston, MA.

Sauer-Budge, A. (2015) Invited Talk. "Rapid sample preparation of viable bacteria directly from blood specimens" Sample Prep 2015. Bethesda, MD.

Sauer-Budge, A. (2015) Invited Talk. "Rapid phenotypic methods for diagnosing infections and antibiotic susceptibility testing." AACC Emerging Clinical & Laboratory Diagnostics: Pushing the Envelope, Los Angeles, CA.

Kalashnikov M, Campbell J, Hanelt I, Lee J, Sharon A, Sauer-Budge AF. (2014) Invited Poster. "Enhanced microfluidic platform for stress-induced rapid antibiotic susceptibility testing." AACC Emerging Clinical & Laboratory Diagnostics Conference, San Jose, CA.

Sauer-Budge, A (2014) Invited talk. "Design for low cost manufacturing of point of care microfluidic devices", SLAS 2014, San Diego, CA.

Sauer-Budge, A (2013) Invited talk. "Rapid Bacterial Sample Preparation from Blood", Sample Prep 2013, San Diego, CA.

Sauer-Budge, A (2013) Invited talk. "Automated on-chip bead-based ELISA", Sample Prep East, Boston, MA.

Sauer-Budge, A (2012) Invited talk. "Innovations at the Intersection of Mechanical Engineering and Life Sciences", Boston University Mechanical Engineering Department Seminar Series, Boston, MA.

Sauer-Budge, A. (2012) Invited talk. "Development of a point-of-care rapid and sensitive bacteremia diagnostic," AACC Oakridge 2012, San Jose, CA.

Sauer-Budge, A (2012) Invited talk. "Isolation of dilute bacteria from blood for rapid diagnostics", Sample Prep 2012, San Diego, CA.

Kalashnikov, M., Campbell, J., Lee, J.C., Sharon, A., Sauer-Budge, A.F. (2012) Poster presentation. "Rapid, Stress-Induced Antibiotic Susceptibility on a Chip". IEEE Micro- and Nano-engineering conference, Maui, HI.

Campbell J, Sauer-Budge AF, and Sharon A. (2012) Poster presentation. "Research at the Intersection of Life Sciences and Engineering: Developing Microfluidic Platforms for Diagnostic Applications". Gordon Research Conference: Bioanalytical sensors, Newport, RI.

Kalashnikov MK, Lee JC, Sharon A, and Sauer-Budge AF. (2012) Poster presentation.

"Microfluidic platform for stress-induced rapid antibiotic susceptibility testing". AACC Oakridge 2012, San Jose, CA.

Sauer-Budge, A (2009) Invited talk. "Microfabricated tools for programming live neural pathways", Neural Restoration Workshop, Washington D.C.

Sauer-Budge, A. (2008) Invited talk. Boston University Biomedical Engineering Department Seminar Series. "Integrating microfluidic sample preparation and molecular diagnostics into a low cost disposable device"

Sauer-Budge, A. (2005) Invited talk to the NIH Viral Load Working Group, NIH, Bethesda, MD, November.

Sauer-Budge, A., Nyamwanda, J., Lubensky, D. K. and Branton, D. (2003). "Progress towards Nanopore-based single molecule sequencing: The Unzipping of Double-Stranded DNA Forced through a Nanopore Measured at the Single Molecule Level." BioMEMS, The Knowledge Foundation, Cambridge, MA.

Sauer, A., Dulay, M.D., and Zare, R. (1996). "Kinetic Studies of Mixed Base PNA-DNA Hybridization with Capillary Electrophoresis and Laser Induced Fluorescence Detection" Johnson Symposium for Organic Chemistry, Stanford University.

Editorships & Editorial Review Boards

Robotics and Computer-Integrated Manufacturing, Editor for Special Issue on "Translational Research - Where Engineering Meets Medicine" (2011).

Peer Reviewer

Journals

Lab On A Chip

Scientific Reports

Bioengineering

PLOS One

Antibiotics

ASME Journal of Nanotechnology in Engineering and Medicine

RSC Advances

NIH Review Panels

NIH Center for Scientific Review Enabling Bioanalytical and Imaging Technologies (EBIT),
Standing member, 2017 - 2021

NIH Center for Scientific Review EBIT, Ad hoc reviewer, 2016

NIH Center for Scientific Review Clinical Research and Field Studies (CRFS), Ad Hoc
Reviewer, 2016

NIH/NIAID ZAI1 RRS-M (C2), Contracts Sample Preparation, 2015

NIH/NIAID ZAI MM-1 (M1), Development of Sample Sparing Assays for Monitoring Immune
Responses, 2015

NIH ZRG1 F130C 20, Center for Scientific Review Special Emphasis panel, Fellowships, 2015

NIH /NIAID ZAI1 ALW0M (J1), NIAID Clinical Trial Planning Grant (R34), 2015

NIH/NIGMS ZGM1 BBCB-A(BI), Biomedical Instrumentation Special Emphasis Panel (SEP),
2014

NIH/NIMH ZMH1 ERB-L (04), BRAIN Initiative Review Special Emphasis Panel (SEP), 2014

NIH Center for Scientific Review CRFS, Ad Hoc Reviewer, 2014

NIH/NIAID ZAI MFH-M (J1), Partnerships for Diagnostics to Address Antimicrobial
Resistance of Select Bacterial Pathogens, 2014

NIH/NIGMS ZGM1 BBCB-A (BT), Biomedical Instrumentation Special Emphasis Panel (SEP),
2013

NIH/NIAID ZAI-LG-M-J1, Partnerships for Biodefense (Diagnostics), 2012

NIH/NHLBI SBIR Contract Proposals: Phase 2, Protein Capture Agents for Cardiovascular
Research, 2011

NIH/NIAID U.S. India Bilateral Collaborative Research Partnerships (CRP) for Prevention of
HIV/AIDS and Co-Morbidities, 2010

NIH/NIAID ZAI1 LG-M (J3) Partnerships for Biodefense (Diagnostics), 2010

NIH/NHLBI SBIR Contract Proposals: Phase 1 Protein Capture Agents for Cardiovascular
Research, 2010

Appendix B: Testimony List

1. *Abad v. Mister East, Amikle Restaurant, Inc., and Central Park Bar Restaurant Sushi, Superior Court of New Jersey Law Division: Union County* in January 2020. I was retained on behalf of First Mercury Insurance and Starr Indemnity & Liability for the insured Mister East.
2. *Mario Badescu Skin Care, Inc. v. Sentinel Insurance Company Limited, United States District Court: Southern District of New York* in June 2021. I was retained on behalf of Sentinel Insurance.
3. *Santa Ynez Band of Chumash Mission Indians of the Santa Ynez Reservation, California v. Lexington Insurance Company, Superior Court of California, County of Santa Barbara* in March 2022. I was retained on behalf of Lexington Insurance Company.

Appendix C: List of Materials Considered

The full list of materials considered includes those referenced specifically in the report and those included in this Appendix B.

Legal Documents

Complaint for Declaratory Relief. United States Bankruptcy Court, District of Delaware, dated December 21, 2020, Case No. 20-11558

Plaintiff's Response to Defendant Allied World National Assurance Company's Requests for Production. United States Bankruptcy Court, District of Delaware, dated July 14, 2021, Case No. 20-11558

Plaintiff's Response to Defendant Continental Casualty Company's First Requests for Admission. United States Bankruptcy Court, District of Delaware, dated July 14, 2021, Case No. 20-11558

Plaintiff's Response to Defendant Continental Casualty Company's First Requests for Production. United States Bankruptcy Court, District of Delaware, dated July 14, 2021, Case No. 20-11558

Plaintiff's Response to Defendant Continental Casualty Company's First Set of Interrogatories. United States Bankruptcy Court, District of Delaware, dated July 14, 2021, Case No. 20-11558

Plaintiff's Response to Defendants Endurance American Specialty Insurance Company; Starr Surplus Lines Insurance Company; Allianz Global Risks US Insurance Company; Liberty Mutual Insurance Company; Certain Underwriters at Lloyd's of London Subscribing to Policy No. WC27C0A190101; Allied World National Assurance Company; QBE Specialty Insurance Company and General Security Indemnity Company of Arizona's First Requests for Production. United States Bankruptcy Court, District of Delaware, dated July 14, 2021, Case No. 20-11558

Plaintiff's Response to Defendants Endurance American Specialty Insurance Company; Starr Surplus Lines Insurance Company; Allianz Global Risks US Insurance Company; Liberty Mutual Insurance Company; Certain Underwriters at Lloyd's of London Subscribing to Policy No. WC27C0A190101; Allied World National Assurance Company; QBE Specialty Insurance Company and General Security Indemnity Company of Arizona's First Set of Interrogatories. United States Bankruptcy Court, District of Delaware, dated July 14, 2021, Case No. 20-11558

Plaintiff's Response to Defendant Allied World National Assurance Company's Second Set of Interrogatories, United States Bankruptcy Court, District of Delaware, dated November 10, 2021, Case No. 20-11558

Allied World National Assurance Company's Response to Plaintiff's First Set of Interrogatories, United States Bankruptcy Court, District of Delaware, dated December 22, 2021, Case No. 20-11558

Defendant Allied World National Assurance Company's Answers and Objections to Plaintiff's Second Set of Interrogatories, United States Bankruptcy Court, District of Delaware, dated July 21, 2022, Case No. 20-11558

Defendant Allied World National Assurance Company's Supplemental Answers and Objections to Plaintiff's Second Set of Interrogatories, United States Bankruptcy Court, District of Delaware, dated November 2, 2022, Case No. 20-11558

Allied World Commercial Property Policy Declarations, Insured to 24 Hour Fitness Worldwide, Inc. Policy number: 0311-9178-1N

Scheduled Location Pollution Liability Policy Declarations, Allied World National Assurance Company. Policy number: 0309-1873

Depositions

Deposition of Mr. Matt Piro (Individual), dated April 27, 2022, Case No. 20-11558, and exhibits

Deposition of Mr. Dan Larson (Individual), dated April 28, 2022, Case No. 20-11558, and exhibits

Deposition of Mr. Dan Larson 30 (b) (6), dated April 28, 2022, Case No. 20-11558, and exhibits

Deposition of Mr. Jeremy Gottlieb, dated June 17, 2022, Case No. 20-11558, and exhibits

Deposition of Ms. Amy Christensen, dated June 24, 2022, Case No. 20-11558, and exhibits

Deposition of Mr. Jason Carter, dated July 21, 2022, Case No. 20-11558, and exhibits

Deposition of Mr. Tony Ueber, dated July 27, 2022, Case No. 20-11558, and exhibits

Deposition of Mr. Matt Piro 30 (b) (6), dated October 5, 2022, Case No. 20-11558, and exhibits

Expert Reports

Plaintiff's Expert Disclosure Pursuant to Federal Rule of Civil Procedure 26(a)(2). United States Bankruptcy Court, District of Delaware, dated October 21, 2022, Case No. 20-11558, Expert report of Mercedes Carnethon, Ph.D.

Produced Documents

CNA-00000037-00000055

CNA-00000083-00000098

CNA-00000073-00000082

CNA-00000691-00000703

CNA-00000060-00000072

CNA-00000529-00000545

CNA-00000546-00000560

CNA-00000561-00000583

CNA-00000683-00000690

CNA-00000174-00000200

Public Documents and Literature

24 Hour Fitness. 2022. Gym Experience. https://www.24hourfitness.com/gym_experience/. Accessed on 11/14/2022.

24 Hour Fitness. 2022. Lakewood Super-Sport Gym. <https://www.24hourfitness.com/gyms/lakewood-ca/lakewood-super-sport>. Accessed on 11/14/2022.

Aboubakr, H. A., *et al.* 2021. Stability of SARS-CoV-2 and other coronaviruses in the environment and on common touch surfaces and the influence of climatic conditions: A review. *Transbound Emerg Dis* 68(2):296-312.

Abraham, J. P., *et al.* 2020. Using heat to kill SARS-CoV-2. *Rev Med Virol* 30(5):e2115.

Adamczyk, Z., *et al.* 2021. SARS-CoV-2 virion physicochemical characteristics pertinent to abiotic substrate attachment. *Curr Opin Colloid Interface Sci* 55:101466.

Ahn, J. Y., *et al.* 2020. Environmental contamination in the isolation rooms of COVID-19 patients with severe pneumonia requiring mechanical ventilation or high-flow oxygen therapy. *J Hosp Infect* 106(3):570-576.

Akter, S., *et al.* 2021. Prevalence and stability of SARS-CoV-2 RNA on Bangladeshi banknotes. *Sci Total Environ* 779:146133.

Alfano, V., and S. Ercolano. 2020. The Efficacy of Lockdown Against COVID-19: A Cross-Country Panel Analysis. *Appl Health Econ Health Policy* 18(4):509-517.

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Appendix D: Summary of Published Stability Studies for SARS-CoV-2

Table 2. Summary of various published stability studies for SARS-CoV-2. Reproduced as a subset of Table 1 from Buekert *et al.* and expanded with additional references (continued on the following pages). Durations of infectivity are presented as ranges, where the lower value corresponds to the final sampling point at which infectious virus could be recovered from the surface and the upper value corresponds to the first sampling point at which the quantity of infectious virus was below the lower limit of detection (LOD). If only a single number, infectious virus was only detected at the first time point, whereas “<” a single number denotes infectious virus was not detected at the first time point and “>” a single number denotes infectious virus was detected at the last timepoint of the experiment. The approximate titer reductions are in units TCID₅₀/mL unless otherwise noted; the first value corresponds to the initial titer and the proceeding value corresponds to the LOD. Media is the matrix used to dry the viruses on the surfaces. Type is the name of the strain or isolate of the virus. Relative humidity: RH

Ref	Substrate	Time	Half-Life (hours)	Titer (log)	Temp (°C)	RH (%)	Media	Virus (Type)					
Van Doremalen <i>et al.</i> ²⁶⁹ (published March 2020)	Plastic	72-96 h	6.81	3.5–0.6	21-23	65	Culture Medium	SARS-CoV-2 (nCoV-WA1-2020)					
	Stainless Steel	48-72 h	5.63										
	Cardboard	24-48 h	3.46										
	Copper	4-8 h	0.77	3.5–1.5				SARS-CoV-1 (Tor 2)					
	Plastic	72-96 h	7.55	3.5–0.6									
	Stainless Steel	48-72 h	4.16										
	Cardboard	8-24 h	0.59	3.5–1.5									
	Copper		1.5										
Chin <i>et al.</i> ²⁷⁰ (published April 2020)	Outer Surgical Mask	>7 d	1.4, 23.9	7.8-2	22	65	Culture medium	SARS-CoV-2 (n/a)					
	Inner Surgical Mask		1.0, 9.9										
	Plastic	4-7 d	1.6, 11.4										
	Stainless steel		0.3, 14.7										
	Glass	2-4 d	1.2, 4.8										
	Banknote		0.9, 7.9										
	Wood, Cloth	1-2 d	n/a										
	Printing/ Tissue Paper	1-3 h											

²⁶⁹ van Doremalen, N., *et al.* 2020. Aerosol and Surface Stability of SARS-CoV-2 as Compared with SARS-CoV-1. N Engl J Med 382(16):1564-1567.

²⁷⁰ Chin, A. W. H., *et al.* 2020. Stability of SARS-CoV-2 in different environmental conditions. Lancet Microbe 1(1):e10.

Ref	Substrate	Time	Half-Life (hours)	Titer (log)	Temp (°C)	RH (%)	Media	Virus (Type)
Harbourt <i>et al.</i> ²⁷¹ (published June 2020)	Swine Skin	>336 h	46.8	4.5-0.1*	4	40-50	Culture Medium	SARS-CoV-2 (USA-WA 1/2020)
		96-168 h	3.5		22			
		8-24 h	0.6		37			
	USD 1 Bank Note (25% Linen and 75% Cotton)	168-336 h	33.2		4			
		8-24 h	1.3		22			
		4-8 h	0.4		37			
	USD 20 Bank Note (25% Linen and 75% Cotton)	168-336 h	15.9		4			
		24-72 h	1.1		22			
		8-24 h	0.6		37			
	Clothing (35% Cotton and 65% Polyester)	96-168 h	33.7		4			
			1.0		22			
		4-8 h	0.2		37			
Kratzel <i>et al.</i> ²⁷² (published June 2020)	Metal	192-214 h	12.9	7.3-2	4	30-40	Culture Medium w/0.3% BSA	SARS-CoV-2 (Munchen-1.1/2020/929)
		120-144 h	9.1		RT			
		>214 h	17.9		30			
Biryukov <i>et al.</i> ²⁷³ (published July/Aug 2020)	Stainless steel, Acrylonitrile Butadiene Styrene Plastic, Nitrile Rubber Gloves (data from all materials were averaged)	>48 h	15.33	2-0.2	24	20	Simulated Saliva	SARS-CoV-2 (USA-WA 1/2020)
		24-48 h	11.52			40		
		24-48 h	9.15			60		
		n/a	8.33			80		
		n/a	6.11	n/a	28	40		
		9-24 h	7.33	2-0.2	35	20		
		3-9 h	7.52			40		
		0.75-3 h	2.26			60		

²⁷¹ Harbourt, D. E., *et al.* 2020. Modeling the stability of severe acute respiratory syndrome coronavirus 2 (SARS-CoV-2) on skin, currency, and clothing. PLOS Neglected Tropical Diseases 14(11):e0008831.

²⁷² Kratzel, A., *et al.* 2020. Temperature-dependent surface stability of SARS-CoV-2. Journal of Infection 81(3):452-482.

²⁷³ Biryukov, J., *et al.* 2020. Increasing Temperature and Relative Humidity Accelerates Inactivation of SARS-CoV-2 on Surfaces. mSphere 5(4):e00441-00420.

Ref	Substrate	Time	Half-Life (hours)	Titer (log)	Temp (°C)	RH (%)	Media	Virus (Type)
Matson <i>et al.</i> ²⁷⁴ (published Sept 2020)	Polystyrene	24-48 h	3.3	5-0.5	4	40	Nasal Mucus	SARS-CoV-2 (USA-WA1/2020)
		8-24 h	3.1		21			
		8-24 h	1.5		27			
		24-48 h	5.8		4	Sputum		
		8-24 h	3.1		21			
		8-24 h	1.5		27			
Pastorino <i>et al.</i> ²⁷⁵ (published Sept 2020)	Polypropylene	>96 h	>96	6-0.5	19-21	45-55	Culture Medium w/1.8 g/L FBS FBS Culture Medium + 10 g/L BSA	SARS-CoV-2 (n/a)
	Glass	24–48 h >96 h	17 >96				Culture Medium w/1.8 g/L FBS FBS Culture Medium + 10 g/L BSA	
	Aluminum	2-4 h >96 h	2.5 >96				Culture Medium w/1.8 g/L FBS FBS Culture Medium + 10 g/L BSA	
Liu <i>et al.</i> ²⁷⁶ (published Oct 2020)	Plastic	>168 h	0.57, 16.38	6-1.5	25-27	35	Culture Medium	SARS-CoV-2 (BetaCoV/Beijing/AMMS01/2020)
	Stainless Steel		0.83, 22.88					
	Glass		0.84, 22.30					
	Surgical mask		0.64, 19.07					
	Ceramics		0.51, 21.71					
	Latex Gloves		0.54, 10.28					
	Wood	0.20, 21.41						
	Cotton Clothes	96-120 h	0.17, 22.72					
	Paper	96-120 h	4.75					

²⁷⁴ Matson, M. J., *et al.* 2020. Effect of Environmental Conditions on SARS-CoV-2 Stability in Human Nasal Mucus and Sputum. *Emerg Infect Dis* 26(9).

²⁷⁵ Pastorino, B., *et al.* 2020. Prolonged Infectivity of SARS-CoV-2 in Fomites. *Emerging Infectious Disease journal* 26(9):2256.

²⁷⁶ Liu, Y., *et al.* 2021. Stability of SARS-CoV-2 on environmental surfaces and in human excreta. *J Hosp Infect* 107:105-107.

Ref	Substrate	Time	Half-Life (hours)	Titer (log)	Temp (°C)	RH (%)	Media	Virus (Type)
Riddell <i>et al.</i> ²⁷⁷ , (published Oct 2020)	Stainless steel	21-28 d	43.2	5-0.8	20	50	2.5 mg/mL BSA, 3.5 mg/mL tryptone and 0.8 mg/mL mucin	SARS-CoV-2 (BetaCoV/Australia/SA01/2020)
		>7 d	12.6		30			
		1 – 24 h	1.5		40			
	Polymer note	>28 d	49.4		20			
		>7 d	14.7		30			
		1-24 h	1.4		40			
	Paper note	>28 d	65.8		20			
		7 -14 d	32.7		30			
		1-24 h	1.6		40			
	Glass	>28 d	45.6		20			
		3 - 7 d	10.5		30			
		1-24 h	2.0		40			
	Cotton	3 – 7 d	40.3		20			
		1 – 3 d	11.0		30			
	Vinyl	> 28 d	48.8		20			
		>3 d	10.1		30			
		1- 2 d	3.0		40			
Hirose <i>et al.</i> (published October 2020) ²⁷⁸	Stainless steel	72-96 h	33	1.0 × 10 ⁵ FFU -n/a	25	45-55	Cell culture medium (DMEM)	SARS-CoV-2
		48-72 h	25	1.0 × 10 ⁵ TCID ₅₀ -n/a	25	45-55	Mucus	SARS-CoV-2
	Borosilicate Glass	72-96 h	33	1.0 × 10 ⁵ FFU -n/a	25	45-55	Cell culture medium (DMEM)	SARS-CoV-2
		48-72 h	24	1.0 × 10 ⁵ TCID ₅₀ -n/a	25	45-55	Mucus	SARS-CoV-2
	Polystyrene	48-72 h	23	1.0 × 10 ⁵ FFU -n/a	25	45-55	Cell culture medium (DMEM)	SARS-CoV-2
		24-48 h	13	1.0 × 10 ⁵ TCID ₅₀ -n/a	25	45-55	Mucus	SARS-CoV-2
	Human skin	6-12 h	3	1.0 × 10 ⁵ FFU -n/a	25	45-55	Cell culture medium (DMEM)	SARS-CoV-2
		6-12 h	4	1.0 × 10 ⁵ TCID ₅₀ -n/a	25	45-55	Mucus	SARS-CoV-2
Magurano <i>et al.</i> ²⁷⁹ (published November 2020)	Polypropylene	84-96 h	n/a	4 - 0.67	20-25	35-45	Cell culture medium (EMEM w/ fetal calf serum)	(SARS-CoV-2) BetaCoV/Italy/CDG1/2020/EPI ISL 412973j2020-02-20
				4 – 0.25	28			

²⁷⁷ Riddell, S., *et al.* 2020. The effect of temperature on persistence of SARS-CoV-2 on common surfaces. *Virology Journal* 17(1):145.

²⁷⁸ Hirose, R., *et al.* 2021. Survival of Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) and Influenza Virus on Human Skin: Importance of Hand Hygiene in Coronavirus Disease 2019 (COVID-19). *Clin Infect Dis* 73(11):e4329-e4335.

²⁷⁹ Magurano, F., *et al.* 2021. SARS-CoV-2 infection: the environmental endurance of the virus can be influenced by the increase of temperature. *Clin Microbiol Infect* 27(2):289 e285-289 e287.

Ref	Substrate	Time	Half-Life (hours)	Titer (log)	Temp (°C)	RH (%)	Media	Virus (Type)
Kasloff <i>et al.</i> ²⁸⁰ (published Jan 2021)	Nitrile Gloves	7-14 d	n/a	7.88-0.5	20	35-40	Tripartite Soil Load w/Mucin, BSA, Tryptone	SARS-CoV-2 (hCoV-19/Canada/ON-VIDO-01)
	Chemical Gloves	4-7 d						
	N-95 Mask	>21 d						
	N-100 Mask, Tyvek Coveralls, Plastic from face shields, stainless steel	14-21 d						
	Heavy Cotton	4 h-1 d						
Gidari <i>et al.</i> ²⁸¹ (published Jan 2021)	Plastic	> 5 d	5.3	4.5- n/a	23-25	40-50	Cell culture medium	SARS-CoV-2 (n/a)
	Stainless steel	2-3 d	4.4					
	Glass	4-5 d	4.2					
Ronca <i>et al.</i> ²⁸² (published Feb 2021)	Acrylic solid surface	24-30 h	n/a	200* - n/a	25	45-50	Cell culture media	SARS-CoV-2 (USA-WA1/2020)
	Solid surface with CuO	not detected						
	Stainless steel, brushed	8-12 h						
	High-pressure laminate	4 -8 h						
	Copper sheet	not detected						
	Quartz	24-30 h						
	Rubber flooring	4-8h						
	Vinyl	8-12 h						
	Wood laminate floor, commercial	4-8 h						
	Luxury vinyl tile	8-12 h						
	Carpet	4-12 h						
	Upholstery, nonwoven	8-12 h						
	Vinyl Wall Covering	48-168 h						

²⁸⁰ Kasloff, S. B., *et al.* 2021. Stability of SARS-CoV-2 on critical personal protective equipment. Scientific Reports 11(1):984.

²⁸¹ Gidari, A., *et al.* 2021. SARS-CoV-2 Survival on Surfaces and the Effect of UV-C Light. Viruses 13(3).

²⁸² Ronca, S. E., *et al.* 2021. SARS-CoV-2 Viability on 16 Common Indoor Surface Finish Materials. Herd:1937586721991535.

Ref	Substrate	Time	Half-Life (hours)	Titer (log)	Temp (°C)	RH (%)	Media	Virus (Type)
Kwon <i>et al.</i> (published Feb 2021) ²⁸³	Nitrile gloves – outer surface	>4 d	11.56	4.7-0.767	21	60	Cell culture medium	SARS-CoV-2 (USA-WA1/2020)
		2-3 d	4.42		25	70		
		7-10 d	22.94		13	66		
		>21 d	85.71		5	75		
	Tyvek	4-5 d	9.36		21	60		
		2-3 d	4.57		25	70		
		>7 d	31.82		13	66		
		>21 d	90.59		5	75		
	N95 mask	4-5 d	9.01		21	60		
		2-3 d	4.4		25	70		
		>7 d	27.77		13	66		
		>21 d	106.37		5	75		
	Cloth	<1 d	3.5		21	60		
		<1 d	2.99		25	70		
		3-5 d	19.94		13	66		
		10-15 d	47.94		5	75		
	Styrofoam	>4 d	9.62		21	60		
		2-3 d	4.75		25	70		
		>7 d	24.67		13	66		
		>21 d	112.91		5	75		
	Cardboard	>4 d	12.86		21	60		
		1-2 d	5.03		25	70		
		>7 d	26.93		13	66		
		>21 d	121.78		5	75		
	Concrete	2-3 d	7.96		21	60		
		1-2 d	2.54		25	70		
		>7 d	17.11		13	66		
		>21 d	80.99		5	75		
	Rubber	3-4 d	11.33		21	60		
		2-3 d	5.03		25	70		
		>7 d	28.27		13	66		
		>21 d	115.74		5	75		
	Glass	3-4 d	9.6		21	60		
		2-3 d	5.58		25	70		
		>7 d	27.34		13	66		
		>21 d	92.03		5	75		

²⁸³ Kwon, T., *et al.* 2021. Environmental Stability of SARS-CoV-2 on Different Types of Surfaces under Indoor and Seasonal Climate Conditions. *Pathogens* 10(2).

Ref	Substrate	Time	Half-Life (hours)	Titer (log)	Temp (°C)	RH (%)	Media	Virus (Type)
Kwon <i>et al.</i> (published Feb 2021) ²⁸⁴ continued from previous page	Polypropylene	>3 d	9.02	4-0.767	21	60	Cell culture medium	SARS-CoV-2 (USA-WA1/2020)
		1-2 d	4.51		25	70		
		>7 d	28.75		13	66		
		>21 d	75.54		5	75		
	Stainless steel	>3 d	7.75		21	60		
		1-2 d	3.41		25	70		
		>7 d	23.46		13	66		
		>21 d	70.06		5	75		
	Galvanized steel	>3 d	6.93		21	60		
		1-2 d	4.19		25	70		
		>7 d	24.22		13	66		
		>21 d	67.21		5	75		
Pottage <i>et al.</i> ²⁸⁵ (published April 2021)	Stainless Steel	4-7 d	n/a	5-0.9	19	57	Cell culture medium	SARS-CoV-2, England 02/2020 HCM/V/052 (EPI ISL 407073 England)
	Stainless Steel	4-7 d		4.3-0.9				SARS-CoV-, HumanCoV19 isolate/England/MIG457/2020 (lineage B.1.1.7 or VOC202012/01)
	Stainless Steel	4-7 d		4.35-0.9				SARS-Cov-2, Human nCoV19 isolate/England/H204661641/2020 (lineage B.1.351 or 20H/501Y.V2) Africa

²⁸⁴ Kwon, T., *et al.* 2021. Environmental Stability of SARS-CoV-2 on Different Types of Surfaces under Indoor and Seasonal Climate Conditions. *Pathogens* 10(2).

²⁸⁵ Pottage, T., *et al.* 2021. A comparison of persistence of SARS-CoV-2 variants on stainless steel. *J Hosp Infect* 114:163-166.

Ref	Substrate	Time	Half-Life (hours)	Titer (log)	Temp (°C)	RH (%)	Media	Virus (Type)
Virtanen <i>et al.</i> ²⁸⁶ (published April 2021)	Finn Raccoon Pelt (Fur Side)	3 – 7 d	n/a	6 – n/a*	RT (~21)	40-75	Cell culture medium	SARS-CoV-2/Finland/1/2020
	Finn Raccoon (Skin Side)	<5 m						
	American Mink Pelt (Fur Side)	3 – 7 d & >10 d [†]						
	American Mink Pelt (Skin Side)	<5 m & 0 - 1 d [†]						
	Blue Fox Pelt (Fur Side)	3 – 7 d						
	Blue Fox Pelt (Skin Side)	5 - 30 m						
	Fake Fur (Fur Side)	30 m – 1 d & 0 - 1 d [†]						
	Fake Fur (Skin/Fabric Side)	30 m – 1 d & 1 – 2 d [†]						
	Petri Dish	3 – 7 d & 5 – 6 d [†]						
	Faux Leather	0 – 1 d						
	Cotton	Not detected						
	Polyester	0 – 1 d						
Morris <i>et al.</i> ²⁸⁷ (July 2021)	Polypropylene	>96 h	26.55	~4-0.5	10	40	DMEM supplemented with 2 mM L-glutamine, 2% fetal 325 bovine serum and 100 units/mL penicillin/streptomycin	SARS-CoV-2 (HCoV-19 nCoV-WA1-2020 (MN985325.1))
		>96 h	14.22		10	65		
		>96 h	13.78		10	85		
		48-72 h	6.43		22	40		
		24-48 h	2.41		22	65		
		48-72 h	7.50		22	85		
		24-48 h	3.43		27	40		
		8-24 h	1.52		27	65		
		24-48 h	2.79		27	85		

²⁸⁶ Virtanen, J., *et al.* 2021. Survival of SARS-CoV-2 on Clothing Materials. *Adv Virol* 2021:6623409.

²⁸⁷ Morris, D. H., *et al.* 2021. Mechanistic theory predicts the effects of temperature and humidity on inactivation of SARS-CoV-2 and other enveloped viruses. *Elife* 10.

Ref	Substrate	Time	Half-Life (hours)	Titer (log)	Temp (°C)	RH (%)	Media	Virus (Type)
Paton <i>et al.</i> ²⁸⁸ (published July 2021)	Stainless Steel	6 – 7 d	n/a	~5.5 – 0.8*	21.5 ± 1	45	Cell culture medium	SARS-CoV-2 (England 02/2020 EPI_ISL_407073)
	Tyvek Coverall	6 – 7 d						
	Disposable Gown	6 – 7 d						
	Surgical Mask	>7 d						
	Bank Note	3 – 5 d						
	Polyester Sport Shirt	< 2.5 h						
	Cotton T-Shirt	3 – 5 d						
Hirose <i>et al.</i> ²⁸⁹ (published Nov 2021)	Plain Paper	48-72 h	2.03-4.06	5 - 0.5	25	40-50	Cell culture medium	SARS-CoV-2 (JPN/TY/WK-521)
	Inkjet Paper	4 – 6 h	0.22-0.44					
	Inkjet Photo Paper	12 – 24 h	0.34-0.68					
Onianwa <i>et al.</i> ²⁹⁰ (published July 2022)	Stainless Steel	168-240 h	n/a	~4.8 – 0.2*	4	>85	Cell culture medium	SARS-CoV-2 Alpha (England/MIG457/2020)
		24-48 h	n/a	~4.8 – 0.2*	24	63		SARS-CoV-2 Alpha (England/MIG457/2020)
		168-240 h	n/a	~4.2 – 0.2*	4	>85		SARS-CoV-2 Delta (HCM/V/078 P2 24MAY2021)
		48-72 h	n/a	~4.2 – 0.2*	24	63		SARS-CoV-2 Delta (HCM/V/078 P2 24MAY2021)

*titer reductions are in units PFU/mL; †replicate experiments;

²⁸⁸ Paton, S., *et al.* 2021. Persistence of Severe Acute Respiratory Syndrome Coronavirus 2 (SARS-CoV-2) Virus and Viral RNA in Relation to Surface Type and Contamination Concentration. *Appl Environ Microbiol* 87(14):e0052621.

²⁸⁹ Hirose, R., *et al.* 2022. Stability of SARS-CoV-2 and influenza virus varies across different paper types. *J Infect Chemother* 28(2):252-256.

²⁹⁰ Onianwa, O., *et al.* 2022. Comparison of Surface Persistence of SARS-CoV-2 Alpha and Delta Variants on Stainless Steel at 4 degrees C and 24 degrees C. *Appl Environ Microbiol* 88(14):e0076422.

Appendix E: Summary of Selected Published Environmental Sampling Studies

Table 3. Selected studies assessing infectious virus on surfaces and air samples.

Setting	Findings	Virus Infectivity?
Hospital (Nebraska) ²⁹¹	>70% of surfaces in patient rooms positive for viral RNA	No infectious virus detected
Hospital (London, England) ²⁹²	Surfaces and air samples frequently positive for viral RNA at high cycle thresholds >30; more likely to be positive in areas closer to patients with COVID-19	No infectious virus detected
Hospital (Florida) ²⁹³	Viable virus isolated from 2 patients with COVID-19 from air samples collected 2–4.8 m away with extremely low viral concentrations of 0.006–0.074 TCID ₅₀ units/mL air	4 of 6 samples were found to contain infectious virus
Hospital (Nebraska) ²⁹⁴	Air samples were taken around 6 patients with COVID-19 and particle size measured; viral growth confirmed from particles <1 µm and 1–4 µm in size	3 of 18 samples were found to contain infectious virus
Hospital (Seoul, Republic of Korea) ²⁹⁵	Viable viruses were identified in 1 of 3 patient rooms on surfaces where the virus could be transmitted by droplets.	8 of 93 samples were found to contain infectious virus, 3 of which were on nasal prong/endotracheal tubes
Households (Utah) ²⁹⁶	SARS-CoV-2 RNA was detected in infected households in 23 of 150 sampled surfaces, but only one of those 23 was found to contain infectious virus.	1 of 150 samples was found to contain infectious virus
Hospital (Quebec City, Canada) ²⁹⁷	100 air samples in room of infected patients were taken and 11 samples had SARS-CoV-2 RNA, but no infectious virus was found.	No infectious virus detected
Hospital (Sweden) ²⁹⁸	Investigation of dispersal through the ventilation system found SARS-CoV-2 viral RNA (both N and E genes) in 7 of 19 room vent openings in COVID-19 ward at first sampling, in none of the 4 ventilation shafts, and in 8 of 9 of main exhaust filters, but no infectious virus was found.	No infectious virus detected.
Car (United States) ²⁹⁹	Air samples were collected inside of a car driven by a SARS-CoV-2 infected patient. All 5 filters tested positive for SARS-CoV-2 RNA.	1 of the 5 filters was found to have infectious virus

²⁹¹ Santarpia, J. L., *et al.* 2020. Aerosol and surface contamination of SARS-CoV-2 observed in quarantine and isolation care. *Scientific Reports* 10(1):12732.

²⁹² Zhou, J., *et al.* 2020. Investigating SARS-CoV-2 surface and air contamination in an acute healthcare setting during the peak of the COVID-19 pandemic in London. *Clinical Infectious Diseases*.

²⁹³ Lednicky, J. A., *et al.* 2020. Viable SARS-CoV-2 in the air of a hospital room with COVID-19 patients. *Int J Infect Dis* 100:476-482.

²⁹⁴ Santarpia, J. L., *et al.* 2021. The size and culturability of patient-generated SARS-CoV-2 aerosol. *Journal of Exposure Science & Environmental Epidemiology*.

²⁹⁵ Ahn, J. Y., *et al.* 2020. Environmental contamination in the isolation rooms of COVID-19 patients with severe pneumonia requiring mechanical ventilation or high-flow oxygen therapy. *J Hosp Infect* 106(3):570-576.

²⁹⁶ Marcenac., *et al.* 2021. Detection of SARS-CoV-2 on Surfaces in Households of Persons with COVID-19. *Int. J. Environ. Res. Public Health*, 18, 8184

²⁹⁷ Dumont-Leblond, N., *et al.* 2020. Low incidence of airborne SARS-CoV-2 in acute care hospital rooms with optimized ventilation. *Emerg Microbes Infect* 9(1):2597-2605.

²⁹⁸ Nissen, K., J., *et al.* 2020. Long-distance airborne dispersal of SARS-CoV-2 in COVID-19 wards. *Sci Rep* 10(1):19589.

²⁹⁹ Lednicky, J. A., *et al.* 2021. Isolation of SARS-CoV-2 from the air in a car driven by a COVID patient with mild illness. *Int J Infect Dis* 108:212-216.

Setting	Findings	Virus Infectivity?
Hospital (Sweden) ³⁰⁰	200 samples collected on different surfaces in different areas of the Infectious Disease ward at the Uppsala University Hospital.	0 of 200 samples collected were found to have infectious virus.
University Student Health Center ³⁰¹ (Florida)	Two air samples were collected in a University Student Health Center	No SARs-CoV-2 infectious virus was detected instead two other infectious respiratory viruses were found.
Households (California and Colorado) ³⁰²	1232 sample were collected inside of household with 1 or more COVID-19-positive people	3 of 1232 samples collected had infectious virus.

³⁰⁰ Krambrich, J., *et al.* 2021. SARS-CoV-2 in hospital indoor environments is predominantly non-infectious. *Virol J* 18(1):109.

³⁰¹ Lednicky, J. A., *et al.* 2020. Collection of SARS-CoV-2 Virus from the Air of a Clinic Within a University Student Health Care Center and Analyses of the Viral Genomic Sequence. *Aerosol Air Qual Res* 20(6):1167-1171.

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